

**ANTIMICROBIAL QUINOLONE DERIVATIVES AND USE OF THE
SAME TO TREAT BACTERIAL INFECTIONS**

5

CROSS-REFERENCE TO RELATED APPLICATION

This application claims the benefit of U.S. provisional Application Serial No. 60/257,904 filed December 21, 2000.

10

FIELD OF THE INVENTION

The present invention relates to substituted quinolone derivatives wherein an oxazolidinone, isoxazolinone, or isoxazoline compound is chemically combined with a quinolone. The present invention also relates to a method of preparing pharmacologically active quinolone derivatives and various intermediates used in the method. The present quinolone derivatives are useful as broad spectrum antimicrobial agents effective against a number of human and veterinary Gram positive and Gram negative pathogens, including the *Staphylococci*, for example *S. aureus*; *Enterococci*, for example *E. faecalis*; *Streptococci*, for example *S. pneumoniae*; *Haemophilus*, for example *H. influenza*; *Moraxella*, for example *M. catarrhalis*; and *Escherichia* for example *E. coli*; *Mycobacteria*, for example *M. tuberculosis*; intercellular microbes, for example *Chlamydia* and *Rickettsiae*; and *Mycoplasma*, for example *M. pneumoniae*, amongst others. The present invention also relates to pharmaceutical compositions containing the quinolone derivatives, to methods of treating a bacterial infection using the quinolone derivatives, and to a process for producing the quinolone derivatives.

20

BACKGROUND OF THE INVENTION

25

The increase in bacterial resistance to existing antibacterial agents is a major clinical problem. Accordingly, there is a need in the art for compounds, compositions, and methods of treating warm-blooded animals that suffer from a

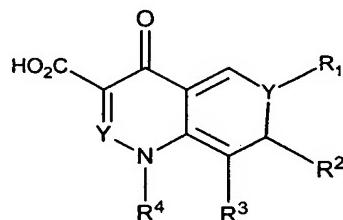
PATENT APPLICATION

28341/6304.NDV1

bacterial infection and are resistant to conventional antibacterial treatments. The development and increase in resistance to the quinolone carboxylic acid class of antibacterial compounds has not been as pervasive as with other antibacterial agents. Therefore, new quinolone carboxylic acid compounds may be useful in combating

5 resistant bacteria.

The 7-substituted quinolone carboxylic acid derivatives, represented by the general formula (II), wherein Y is either C-R⁵ or N, and R¹ through R⁵ include a wide variety of substituents, are well known as anti-fungal and anti-bacterial agents, 10 and as synthetic intermediates to related compounds. The 7-substituted derivatives of compound (II) include the antibacterials cinoxacin (U.S. Patent No. 3,669,965); ciprofloxacin (U.S. Patent Nos. 4,563,459 and 4,620,007); ofloxacin (U.S. Patent No. 4,382,892); and levofloxacin (U.S. Patent Nos. 4,985,557, 5,053,407, and 5,142,046).

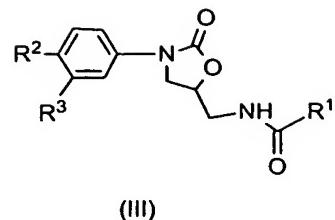


(II)

15

Oxazolidinones having a general structural formula (III) also are a well known class of orally active, synthetic antibacterial agents. The literature contains numerous references to oxazolidinones (III), wherein R¹ through R³ include a wide 20 variety of substituents. Oxazolidinones having one or two substituents on the phenyl ring are disclosed in U.S. Patent Nos. 4,705,799; 5,523,403; and 5,654,435, for example. Oxazolidinones (III) include the antibacterial agent designated as DuP 721, see *J. Med. Chem.*, 32, 1673 (1989).

PATENT APPLICATION
28341/6304.NDV1

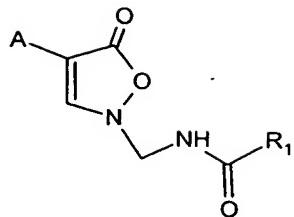


(III)

Oxazolidinones (III) having an arylbenzene substituent on the oxazolidinone ring are disclosed in U.S. Patent Nos. 4,948,801 and 5,130,316. 3-[(Di- or fused-ring substituted)phenyl]-2-oxazolidinones are disclosed in U.S. Patent Nos. 4,977,173; 4,921,869; 4,801,600; and 5,164,510. European Patent Applications 0 697 412; 0 694 544; 0 694 543; and 0 693 491, and International Patent Publication No. WO 93/09103, disclose 5- to 9-membered substituted aryl- and heteroaryl-phenyl oxazolidinones as antibacterial agents. U.S. Patent No. 5,254,577 discloses 10 aminomethyloxooxazolidinyl arylbenzene derivatives as antibacterial agents. Other references disclosing oxazolidinones include U.S. Patent Nos. 4,801,600 and 4,921,869. Some of the pyridine-substituted phenyl oxazolidinone derivatives disclosed in the above patents are effective against Gram positive bacteria, such as *Staphylococcus aureus* and *Streptococcus pneumoniae*. However, the oxazolidinones 15 are not active against Gram negative bacteria, such as *Escherichia coli*, *Klebsiella*, *Proteus*, and *Serratia marcescens*. Moreover, oxazolidinones cannot be administered as an injection solution because their free amino forms are sparingly soluble.

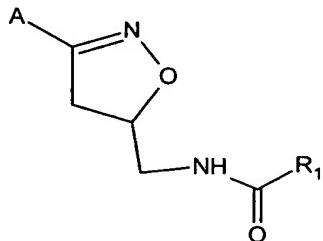
Isoxazolinone derivatives having a general structural formula (IV) are 20 disclosed in WO 00/10566 as anti-bacterial agents. Simple isoxazolinones also are used as preemergent herbicides. For example, U.S. Patent No. 4,065,463 discloses 2-methyl-4-(chloro-m-tolyl)-3-isoxazolin-5-one, which is useful in preventing weed growth.

PATENT APPLICATION
28341/6304.NDV1



(IV)

Isoxazoline derivatives having a general structural formula (V) are disclosed in WO 99/41244, WO 98/07708 and U.S. Patent No. 3,769,295 as anti-microbial agents. Simple isoxazolines also are used as preemergent herbicides. In addition, U.S. Patent No. 4,283,403 discloses 3-aryl-2-isoxazolines, which are useful in preventing plant fungal diseases.



(V)

Although quinolones, oxazolidinones, isoxazolinones, and isoxazolines are known, applicants are aware of no reference which discloses covalently bonding a quinolone to an oxazolidinone, an isoxazolinone, or an isoxazoline, and using the resulting quinolone derivatives as broad spectrum anti-bacterial agents against both Gram positive and Gram negative bacteria.

The present invention is directed to structurally novel compounds produced by covalently bonding an antibacterial oxazolidinone, isoxazolinone, or isoxazoline compound to a substituted quinolone compound. The compounds of the present invention have a quinolone structure substituted with an oxazolidinone,

PATENT APPLICATION

28341/6304.NDV1

isoxazolinone, or isoxazoline via a linking group at the 1- or 7- position of the quinolone.

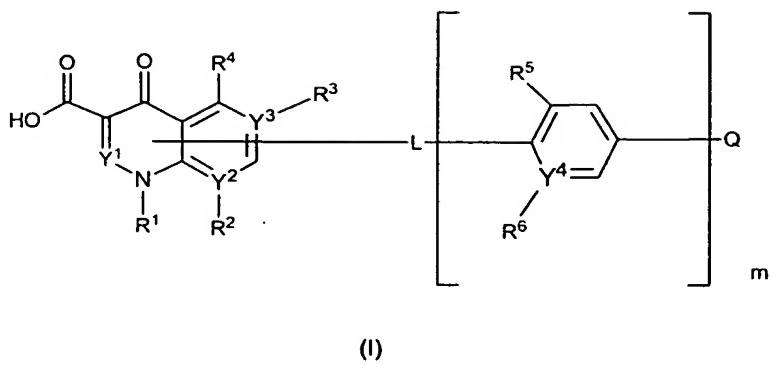
The present compounds are active against Gram negative bacteria and
5 Gram positive bacteria, and accordingly are useful as broad spectrum antibacterial agents. The present compounds are surprisingly effective against a number of human and veterinary pathogens, including *Staphylococci*, for example *S. aureus*; *Enterococci*, for example *E. faecalis*; *Streptococci*, for example *S. pneumoniae*; *Haemophilus*, for example *H. influenza*; *Moraxella*, for example *M. catarrhalis*; and
10 *Escherichia* for example *E. coli*. Other examples include *Mycobacteria*, for example *M. tuberculosis*; intercellular microbes, for example *Chlamydia* and *Rickettsiae*; and *Mycoplasma*, for example *M. pneumoniae*.. The present compounds also are envisioned as cytotoxic anticancer agents.

15 Syntheses of simple quinolone derivatives are well known in the art. However, the synthesis of a quinolone covalently linked to an oxazolidinone, an isoxazolinone, or an isoxazoline is not straightforward. Thus, a method of synthesizing compounds of the present invention also is disclosed.

20 **SUMMARY OF THE INVENTION**

One aspect of the present invention is to provide substituted quinolone derivatives having a structural formula (I):

PATENT APPLICATION
28341/6304.NDV1



or a pharmaceutically acceptable salt, hydrate, or prodrug thereof,

wherein Y^1 is CH or N;

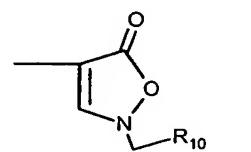
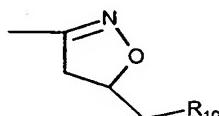
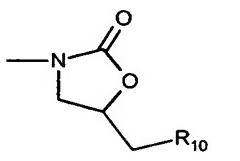
Y^2 , Y^3 , and Y^4 , independently, are C or N;

5 L is a bond or is a linker group attached to a carbon at the seven quinolone ring position or to an N at the one quinolone ring position, and selected from the group consisting of a bond, NR^7 , and $NR^8(CR^9_2)_nNR^8$;

 m is 0 or 1;

 n is 0-3;

10 Q is selected from the group consisting of



R^1 is selected from the group consisting of null, H, C_1-C_4 alkyl, C_3-C_5 cycloalkyl, C_1-C_4 haloalkyl, and halophenyl;

15 R^2 is null when Y^2 is N, or is selected from the group consisting of H, alkyl, C_1-C_2 alkoxy, halo, and haloalkoxy, when Y^2 is C, or when Y^2 is C, R^1 and R^2 can be taken together to form a 5- or 6-membered, optionally substituted, heteroalkyl or heteroaryl ring;

R^3 is H or F when Y^3 is C, or R^3 is null when Y^3 is N;

20 R^4 is selected from the group consisting of H, methyl, amino, and F; R^5 is selected from the group consisting of H, methyl, hydroxy, and halo;

PATENT APPLICATION
28341/6304.NDV1

R⁶ is selected from the group consisting of H, methyl, hydroxy, and halo, when Y⁴ is C, or R⁶ is null when Y⁴ is N;

R⁷ is selected from the group consisting of H, C₁-C₄ alkyl, formyl, alkylcarbonyl, alkylsulfonyl, and alkoxycarbonyl;

R⁸, independently, are H or C₁-C₄alkyl, or are taken together to form a 4- to 9-membered, optionally substituted, heteroalkyl or heteroaryl ring;

R⁹, independently, are H or C₁-C₄alkyl, or are taken together to form a 4- to 9-membered heterocyclic or heterobicyclic ring, optionally substituted with C₁-C₂alkyl, haloalkyl, or methoximino;

R¹⁰ is selected from the group consisting of OH, alkoxy, aryloxy, and NHC(=Z)R¹¹;

R¹¹ is selected from the group consisting of H, C₁-C₇alkyl, C₃-C₅cycloalkyl, hydroxymethyl, haloalkyl, CH₂SMe, NR¹², C₁-C₄alkoxy, and aryloxy;

R¹² is C₁-C₄alkyl; and

Z is O or S.

Another aspect of the present invention is to provide a pharmaceutical composition containing a compound of formula (I) and a pharmaceutical acceptable carrier, diluent, or excipient.

One other aspect of the present invention is to provide a method of treating microbial infections in a mammal comprising administering to the mammal a pharmaceutically effective amount of a compound of formula (I).

Another aspect of the present invention is to provide method of treating a cancer comprising administering to a mammal in need thereof a pharmaceutically effective amount of a compound of formula (I).

Yet another aspect of the present invention is to provide a method of manufacturing a compound of structural formula (I).

**DETAILED DESCRIPTION OF THE
PREFERRED EMBODIMENTS**

5 As used herein, the terms and phrases have the meanings and definitions known in the art. Some of the more commonly used phrases are described in more detail below.

10 “Alkyl” refers to a cyclic, branched, or straight chain chemical group containing only carbon and hydrogen atoms, for example methyl, pentyl, and adamanyl. Alkyl groups can be unsubstituted or substituted with one or more substituents, *e.g.*, halogen, alkoxy, acyloxy, amino, hydroxyl, mercapto, carboxy, benzyloxy, phenyl, and benzyl. Alkyl groups can be saturated or unsaturated (*e.g.*, containing alkenyl or alkynyl subunits), at one or several positions. Typically, alkyl groups contain 1 to about 12 carbon atoms, for example 1 to about 10, or 1 to about 8-15 carbon atoms.

20 “Heteroalkyl” refers to a cyclic, branched, or straight chain chemical group containing carbon, hydrogen and at least one heteroatom. Heteroalkyl includes bicyclic compounds. The heteroatom typically is nitrogen, oxygen, or sulfur. Heteroalkyl groups can be unsubstituted or substituted with one or more substituents, *e.g.*, halogen, alkoxy, acyloxy, amino, hydroxyl, mercapto, carboxy, benzyloxy, phenyl, and benzyl. When the heteroalkyl group contains a nitrogen atom, the nitrogen atom can be primary, secondary, tertiary, or quaternary, or can be in various forms such as an amide or sulfonamide. Heteroalkyl groups can contain one or more unsaturated (*e.g.*, alkenyl or alkynyl) subunits. Typically, heteroalkyl groups contain 1 to about 12 atoms, for example 1 to about 8, or 1 to about 4 carbon atoms.

30 “Aryl” refers to a monovalent aromatic carbocyclic group having a single ring (*e.g.* phenyl), multiple rings (*e.g.* biphenyl), or multiple condensed rings (*e.g.* naphthyl or anthryl). Aryl groups can be unsubstituted or substituted with

PATENT APPLICATION

28341/6304.NDV1

amino, hydroxyl, alkyl, heteroalkyl, alkoxy, halo, mercapto, and other substituents.

Typically, the aryl group is a substituted single ring compound. For example, the aryl group is a substituted phenyl ring.

5

“Heteroaryl” refers to a monovalent aromatic group having a single ring (e.g., pyridyl or furyl) or multiple condensed rings (e.g., indolizinyl or benzothienyl) containing carbon atoms and having at least one heteroatom within the ring. The heteroatom preferably is nitrogen, oxygen or sulfur. Heteroaryl groups can be optionally unsubstituted or substituted with amino, hydroxyl, alkyl, heteroalkyl, alkoxy, halo, mercapto, and other substituents. In one embodiment, the heteroaryl group is substituted pyridyl.

The term “halo” or “halogen” is defined herein to include fluorine, bromine, chlorine, and iodine.

10

The term “haloalkyl” is defined herein as an alkyl group substituted with one or more halo substituents, either fluoro, chloro, bromo, iodo, or combinations thereof. Similarly, “halocycloalkyl” is defined as a cycloalkyl group having one or more halo substituents.

15

The term “alkoxy” and “aryloxy” are defined as -OR, wherein R is alkyl or aryl, respectfully.

The term “hydroxy” is defined as -OH.

20

The term “amino” is defined as -NR₂, wherein each R, independently, is alkyl or hydrogen.

The term “alkylcarbonyl” is defined as R-C(=O)-, where R is alkyl.

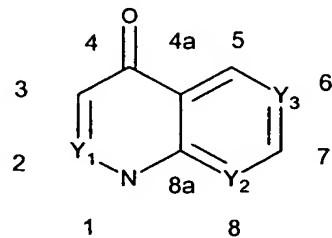
25

The term “alkoxycarbonyl” is defined as RO-C(=O)-, where R is alkyl.

PATENT APPLICATION
28341/6304.NDV1

The term "alkylsulfonyl" is defined as R-SO₃, where R is alkyl.

The quinolone ring system is numbered as follows:

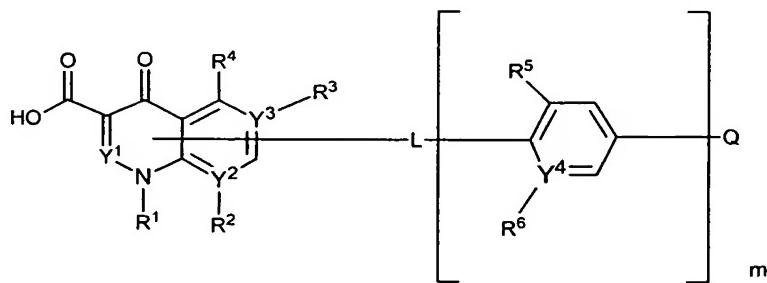


5

"Biologically active compounds" or "bioactive compounds" refers to present quinolone derivatives that exhibit biological activity. For instance, a biologically active compound can inhibit the interaction between an enzyme or receptor and its respective substrate(s) or endogenous ligand(s), or inhibit cell growth of a microorganism, by about at least 15% at a solution concentration of 10⁻³ molar or lower (i.e., has inhibitory activity). For example, a biologically active compound can inhibit such processes at solution concentrations of about 10⁻⁴ M or lower, preferably 10⁻⁵ M or lower, and more preferably about 10⁻⁶ M or lower.

10

The present invention is directed to quinolone-oxazolidinones, quinolone-isoxazolinones, and quinolone-isoxazolines of structural formula (I) as defined below:



(I)

20 or a pharmaceutically acceptable salt, hydrate, or prodrug thereof,

PATENT APPLICATION

28341/6304.NDV1

wherein Y¹ is CH or N;

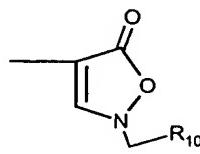
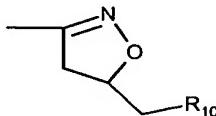
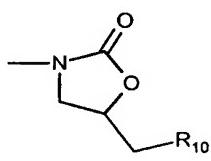
Y², Y³, and Y⁴, independently, are C or N;

L is a bond or is a linker group attached to a carbon at the seven quinolone ring position or to an N at the one quinolone ring position, and selected from the group consisting of a bond, NR⁷, and NR⁸(CR⁹)_nNR⁸;

5 m is 0 or 1;

n is 0-3;

Q is selected from the group consisting of



10 R¹ is selected from the group consisting of null, H, C₁-C₄alkyl, C₃-C₅cycloalkyl, C₁-C₄haloalkyl, and halophenyl;

R² is null when Y² is N, or is selected from the group consisting of H, alkyl, C₁-C₂alkoxy, halo, and haloalkoxy, when Y² is C, or when Y² is C, R¹ and R² can be taken together to form a 5- or 6-membered, optionally substituted, heteroalkyl or heteroaryl ring;

15 R³ is H or F when Y³ is C, or R³ is null when Y³ is N;

R⁴ is selected from the group consisting of H, methyl, amino, and F; R⁵ is selected from the group consisting of H, methyl, hydroxy, and halo;

R⁶ is selected from the group consisting of H, methyl, hydroxy, and halo, when Y⁴ is C, or R⁶ is null when Y⁴ is N;

20 R⁷ is selected from the group consisting of H, C₁-C₄ alkyl, formyl, alkylcarbonyl, alkylsulfonyl, and alkoxy carbonyl;

R⁸, independently, are H or C₁-C₄alkyl, or are taken together to form a 4- to 9-membered, optionally substituted, heteroalkyl or heteroaryl ring;

25 R⁹, independently, are H or C₁-C₄alkyl, or are taken together to form a 4- to 9-membered heterocyclic or heterobicyclic ring, optionally substituted with C₁-C₂alkyl, haloalkyl, or methoximino;

PATENT APPLICATION
28341/6304.NDV1

R¹⁰ is selected from the group consisting of OH, alkoxy, aryloxy, and NHC(=Z)R¹¹;

R¹¹ is selected from the group consisting of H, C₁-C₇alkyl, C₃-C₅cycloalkyl, hydroxymethyl, haloalkyl, CH₂SMe, NR¹², C₁-C₄alkoxy, and aryloxy;

5 R¹² is C₁-C₄alkyl; and

Z is O or S.

The compounds of the present invention are effective antimicrobial agents against a number of human and veterinary pathogens, including Gram-
10 positive, Gram negative, and anaerobic bacteria, and in treating microbial infections in mammals. The present compounds also can be used as cytotoxic anticancer compounds.

Preferred compounds of general formula (I), are those wherein:

15 Y¹ is CH;

Y², Y³, and Y⁴ are C;

L is a bond or NR⁸(CR⁹)_nNR⁸;

n is 2;

R³ is H or F;

20 R⁴ is H, methyl, amino, or F;

R⁵ and R⁶, independently, are H, methyl, hydroxy, or halo;

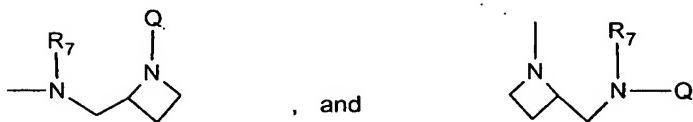
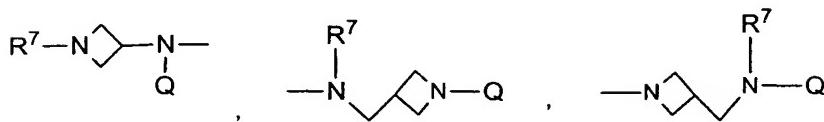
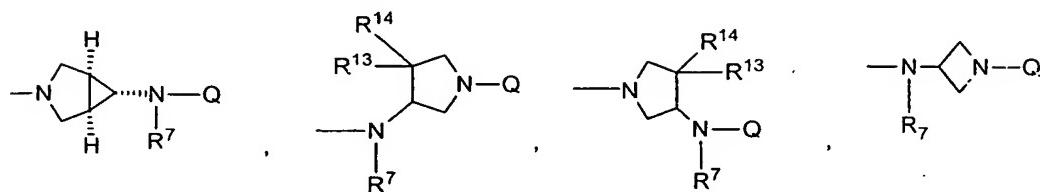
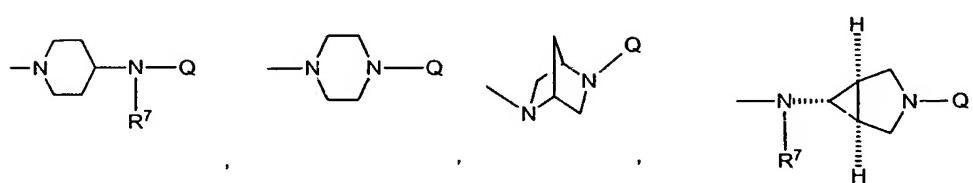
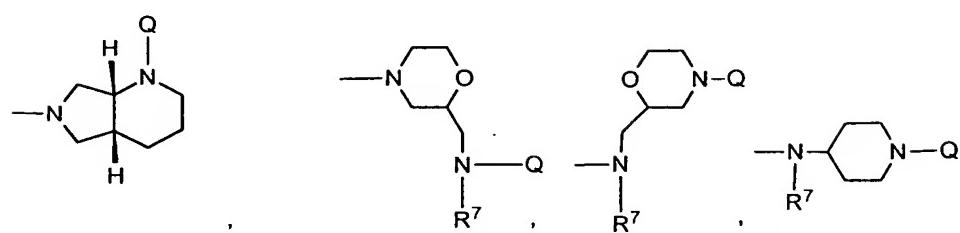
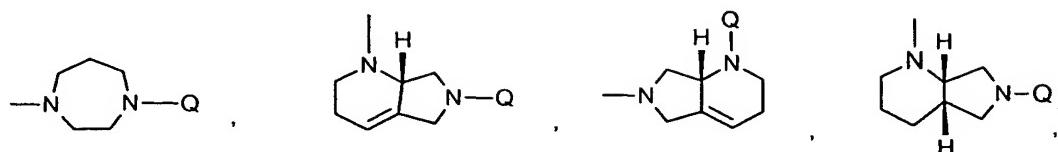
R¹⁰ is OH, alkoxy, aryloxy, or NHC(Z)R¹¹ when Q is an oxazolidinone or isoxazoline group, or is aryloxy, NHC(Z)R¹¹, when Q is a isoxazoline group; and

Z is O.

25

A preferred Q is an oxazolidinone group. Preferred L-Q groups, wherein m is zero, are selected from the group consisting of:

PATENT APPLICATION
28341/6304.NDV1



PATENT APPLICATION

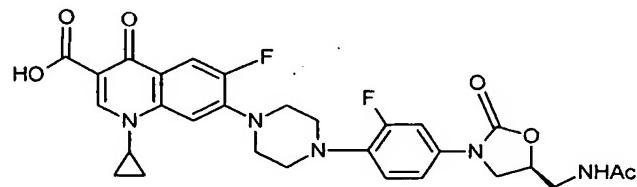
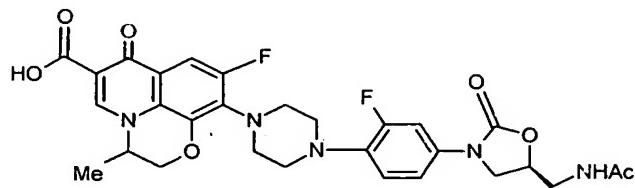
28341/6304.NDV1

wherein R¹³ and R¹⁴, independently, are H, C₁₋₂alkyl, or C₁₋₂ haloalkyl, or are taken together to form a cyclopropyl or methoximino group.

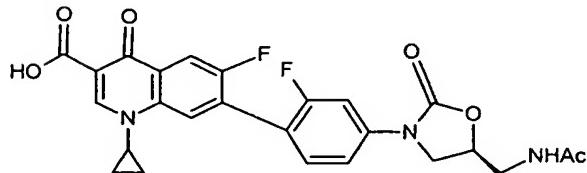
It also is preferred that compounds of formula (I) are optically pure
5 enantiomers having the S-configuration at the five-position carbon of the oxazolidinone or isoxazoline ring.

Preferred compounds of the present invention include (Ac is C(=O)CH₃):

10



and



15

The present invention is also directed to methods of synthesizing pharmaceutically active oxazolidinone-, isoxazoline-, and isoxazolinone- substituted quinolones (I) and intermediate compounds used in the synthesis. The substituted

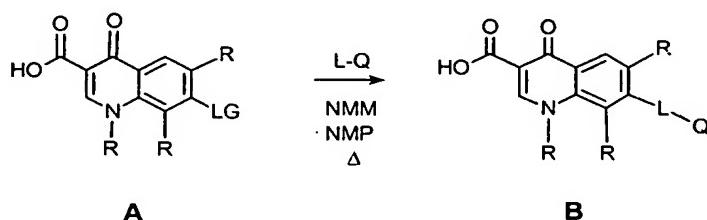
PATENT APPLICATION

28341/6304.NDV1

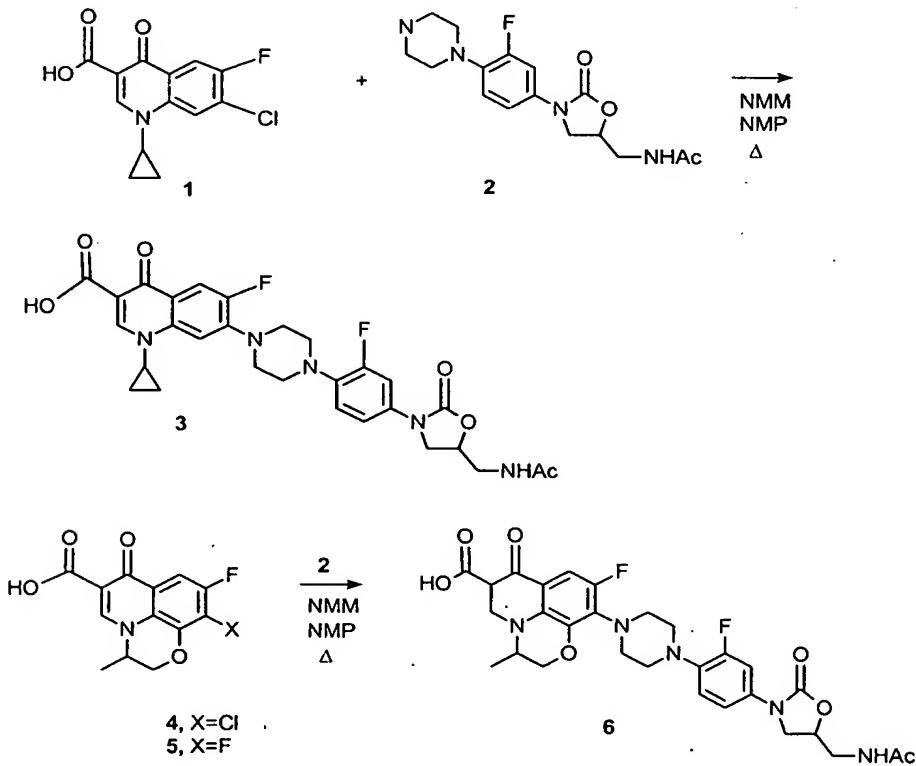
quinolone derivatives of the present invention can be prepared by the following general synthetic schemes.

Scheme 1 illustrates one general method of synthesizing the 7-oxazolidinone-, isoxazoline-, and isoxazolinone- substituted quinolone compounds (B) of the present invention. Scheme 2 illustrates specific examples of synthesizing oxazolidinone-substituted quinolone compounds (3 and 6) of the present invention.

Scheme 1



10 Scheme 2



PATENT APPLICATION
28341/6304.NDV1

In Scheme 1, an appropriately substituted quinolone, preferably containing a leaving group (LG) at the 7-position (compound A), such as a fluoro, chloro, or triflate derivative, is used as a starting material. Specific examples of such compounds are illustrated by compounds (1), (4), and (5). Compounds (1), (4), and (5) are readily available from a number of commercial sources or, alternatively, are known in the chemical literature or can be readily prepared by one skilled in the art.

5 7-Chloro-1-cyclopropyl-6-fluoro-4-oxohydroquinoline-3-carboxylic acid (1) is commercially available from Acros Organics, and its synthesis is described in German Patents DE 3142854, DE 3248505, and DE 3248507. 1-Cyclopropyl-6,7-difluoro-4-

10 oxo-3-quinoliniccarboxylic acid is commercially available from Louston International, and its synthesis is described in German patent DE3248507. 9,10-Difluoro-3-methyl-7-oxo-2,3-dihydro-7H-[1,4]oxazino[2,3,4-λ]quinoline-6-carboxylic acid is commercially available from Maybridge Chemical Company and its synthesis is described in Japanese patents JP 57088182 and JP 58072589 and EP 47005. 9-

15 Chloro-10-fluoro-3-methyl-7-oxo-2,3-dihydro-7H-[1,4]oxazino[2,3,4-λ]quinoline-6-carboxylic acid is commercially available from Commercially available from Zhejiang Hengdian Imp. & Exp. Co., Ltd. and its synthesis is described in Chem. Pharm. Bull., 32, 4907-13 (1984) and EP 206283.

20 In one embodiment, the appropriately substituted quinolone (1), (4), or (5) is treated with an oxazolidinone, isoxazoline, or isoxazolinone substituted with an sufficiently nucleophilic linking group, L, such that the subsequent nucleophilic substitution reaction provides, in a one-pot reaction sequence, the respective crude oxazolidinone-, isoxazoline-, or isoxazolinone-substituted quinolone (I).

25 The L group on the oxazolidinone, isoxazoline, or isoxazolinone can be introduced by standard synthetic methods from commercially available reagents as described hereafter. For example, when quinolone (1), (4), or (5) is treated with 5-(S)-aminomethyl-3-(3-fluoro-4-piperazinophenyl)oxazolidine-2-one in N-

30 methylpyrrolidine-2-one (NMP) and N-methylmorpholine (NMM), the respective crude oxazolidinone-substituted quinolone (3) or (6) is formed in moderate to high yield.

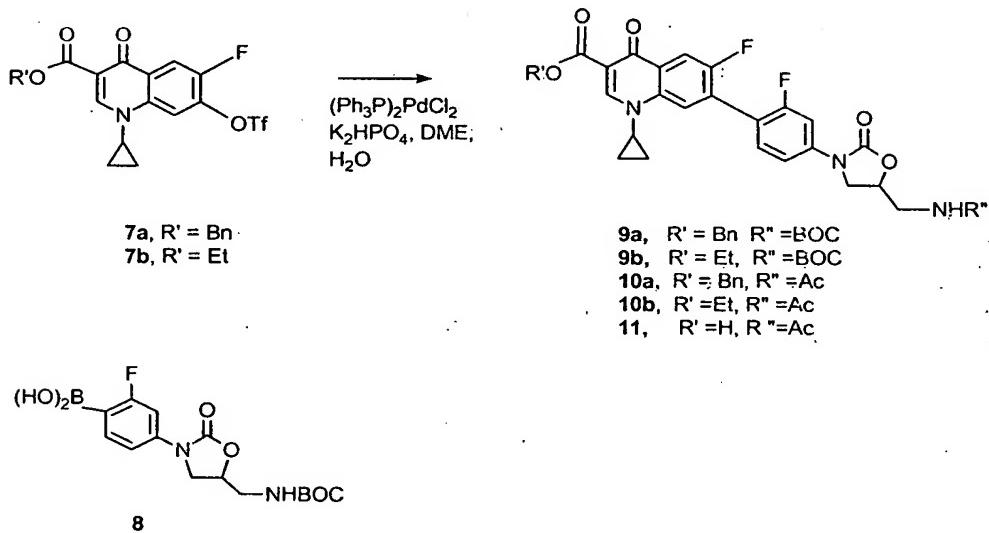
PATENT APPLICATION

28341/6304.NDV1

Compounds (3) and (6) then can be purified following chromatographic techniques well known in the art.

Alternately, Scheme 3 outlines a representative procedure for preparing compounds where a carbon-carbon bond connects the quinolone fragment to a phenyloxazolidinone subunit. Quinolone triflates 7a,b, (Kiely et. al. *J.*

Scheme 3.



10

Heterocyclic Chem. 1991, 28, 1581-1585), is reacted with boronic acid 8 in the presence of 1,2-dimethoxyethane, aqueous dibasic potassium phosphate and a suitable palladium catalyst, such as bis(triphenylphosphine)palladium bichloride or tetrakis(triphenylphosphine)palladium, and at a suitable temperature, preferably at reflux, to generate the respective coupled products 9a,b. It will be apparent to one skilled in the art that compounds 9a,b are both antimicrobial compounds and synthetic intermediates. For example, the *tert*-butoxycarbonyl (BOC) moiety of 9a,b can be removed with, for example, trifluoroacetic acid to give an amino intermediate which can be further elaborated, for example, acylated, employing conditions described below. Additionally, the ester moiety of 10 or a subsequent acylated derivative can be hydrolyzed under acidic or basic conditions to give the

PATENT APPLICATION

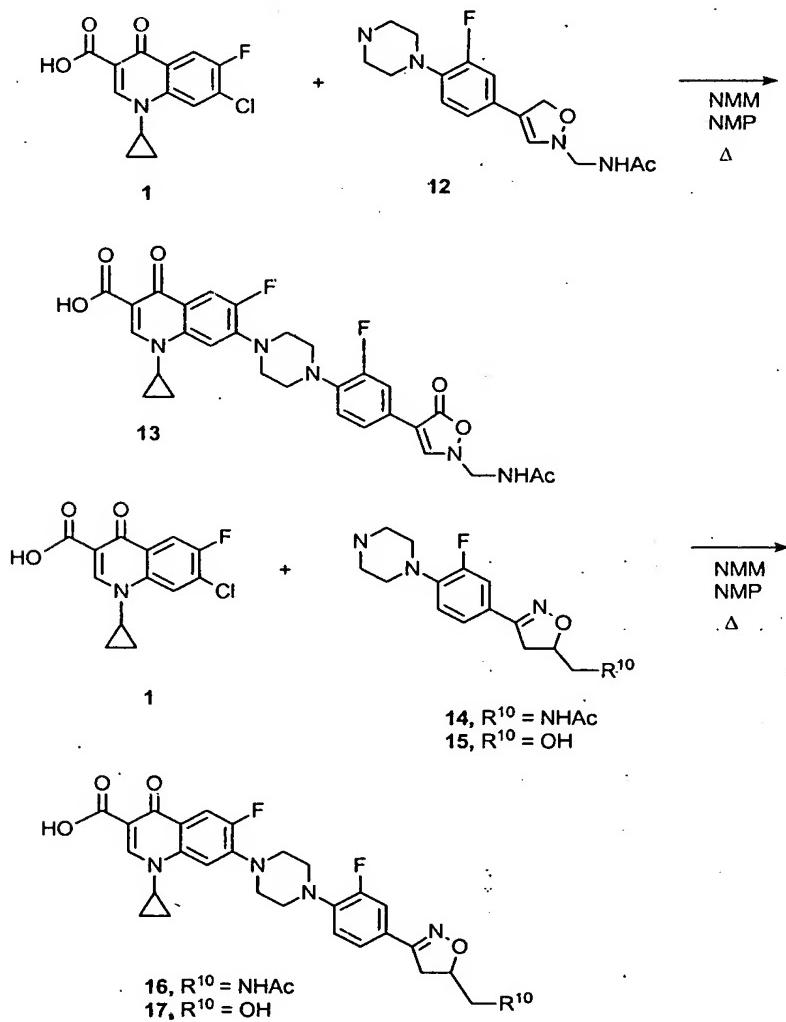
28341/6304.NDV1

corresponding carboxylic acid 11. Furthermore, when R¹ = Bn hydrogenolysis in the presence of a suitable catalyst such as palladium on carbon also affords the corresponding carboxylic acid 11.

As shown in Scheme 4 below, the synthetic procedures leading to isoxazoline- and isoxazolinone- substituted quinolones of the present invention closely parallel the above procedure.

Scheme 4.

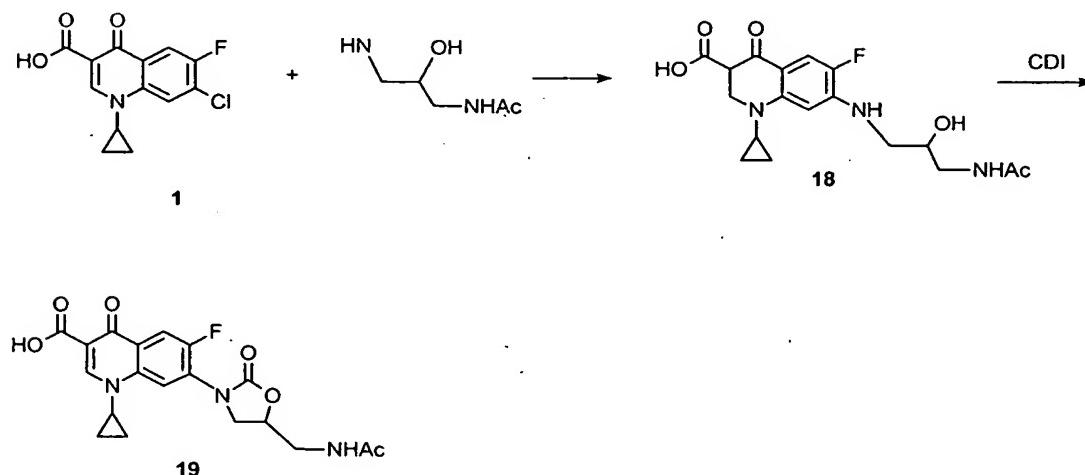
10



PATENT APPLICATION
28341/6304.NDV1

In another embodiment, the linking group between the oxazolidinone, isoxazoline, or isoxazolinone ring and the quinolone ring of the quinolone compounds (Scheme 1) of the present invention is a bond. In these cases, the oxazolidinone, isoxazoline, and isoxazolinone is formed subsequent to the nucleophilic substitution reaction. This is illustrated in Scheme 5.

Scheme 5.



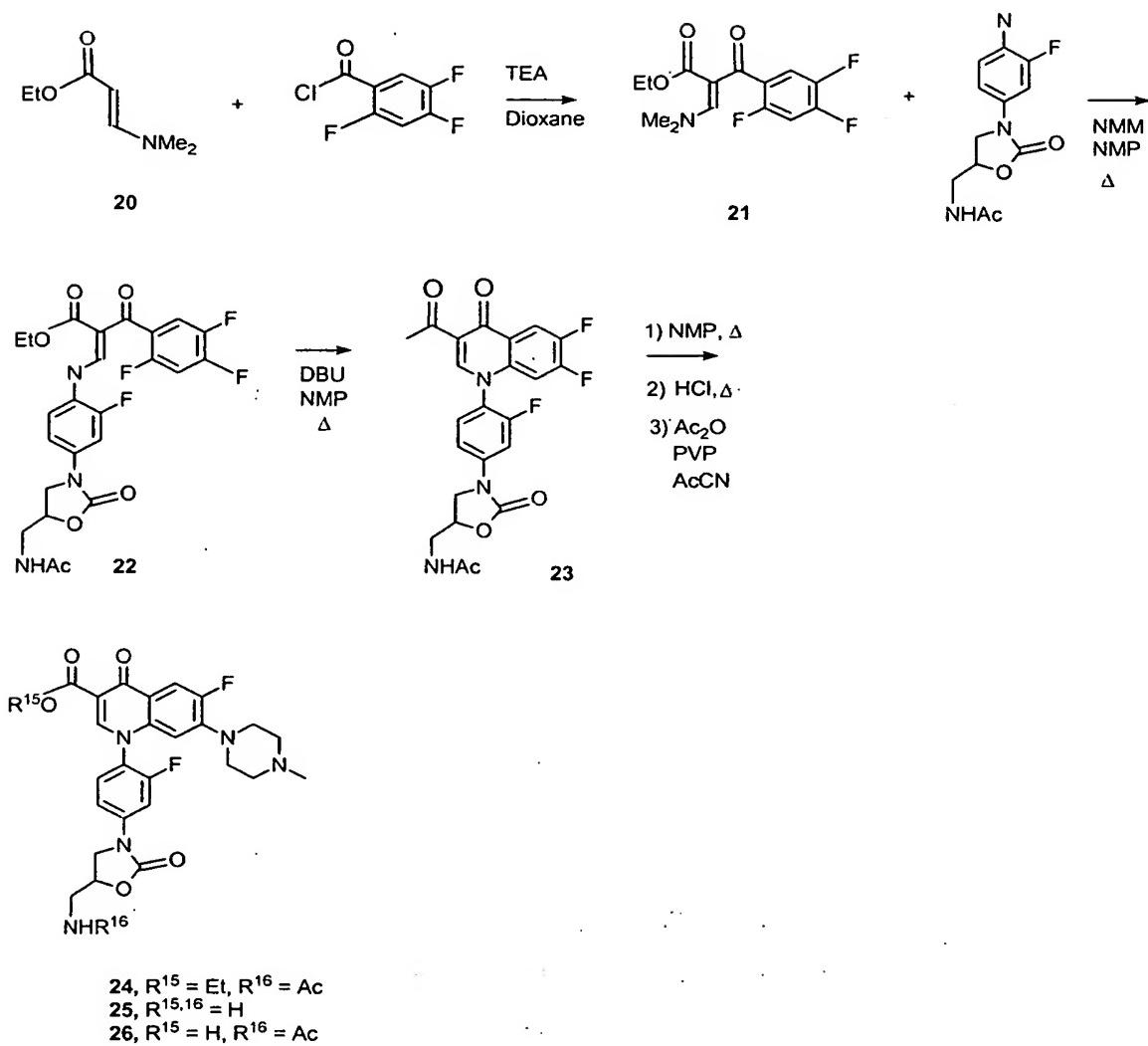
10

For example, quinolone (1) is treated with 1-acetyl-3-amino-2-(S)-oxypropylamine in NMP and NMM, as described above, to form an aminoalcohol intermediate (18). The aminoalcohol intermediate (18) then is converted, in a one-pot reaction sequence, to a crude oxazolidinone-substituted quinolone (19) by treatment with carbonyldiimidazole (CDI). Conversion of amino alcohols to oxazolidinones is achieved by known process as (See e.g., *J. Med. Chem.*, 32, 1673 (1989).).

In another embodiment, the oxazolidinone-, isoxazoline-, and isoxazolinone-substituent is covalently bonded to the one-position nitrogen of the quinolone ring system. Scheme 6 illustrates the synthesis of a 1-oxazolidinone-substituted quinolone compound of the present invention.

PATENT APPLICATION
28341/6304.NDV1

Scheme 6.



5

Briefly, ethyl 3-dimethylaminoacrylate (20) is converted to ethyl 2-(dimethylamino)methylene-2-(2,4,5-trifluorobenzoyl)acetate (21) in the presence of 2,4,5-trifluorobenzoyl chloride, triethylamine, and 1,4-dioxane. Reaction of compound (21) with 5-(S)-acetamidomethyl-3-(4-amino-3-fluorophenyl)-oxazolidine-2-one provides compound (22), which then is heated in the presence of diazabicyclo[4.4.0]undec-2-ene (DBU) and an excess of NMP to facilitate cyclization

10

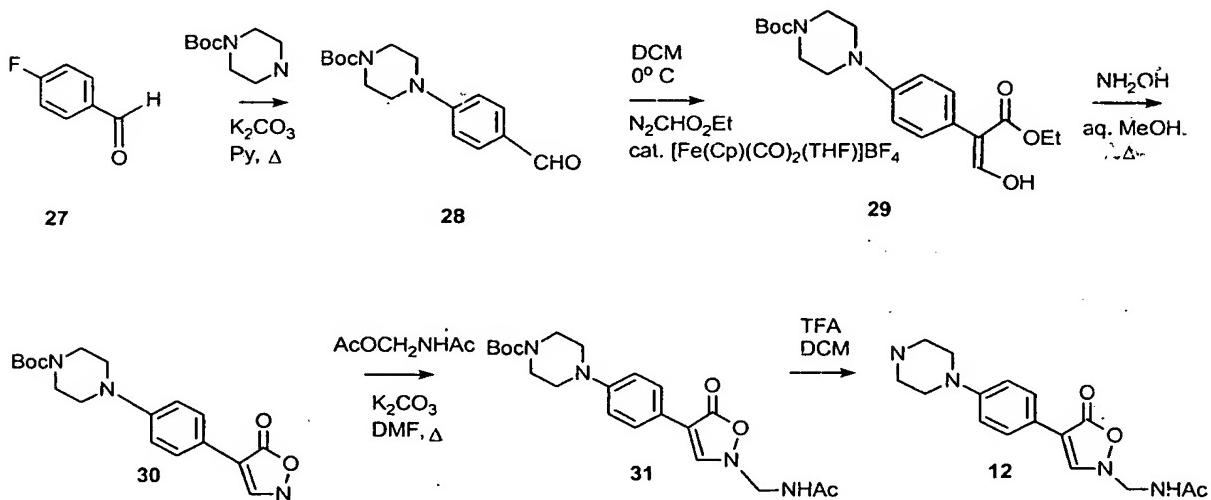
PATENT APPLICATION

28341/6304.NDV1

to quinolone-oxazolidinone (23). Quinolone-oxazolidinone (23) can be converted directly to the corresponding N-methylpiperazinyl compound (24) by treatment with 1-methylpiperazine in the presence of NMP. Hydrolysis of compound (24) in the presence of aqueous hydrogen chloride yields acid amine (25). Acylation of amine (25) provides a oxazolidinone-substituted quinolone (26) of the present invention. The synthetic procedures which provide the N¹-isoxazoline- and isoxazolinone-substituted quinolones of the present invention closely parallel the above procedure.

Isoxazoline- and isoxazolinone intermediates (12), (14), and (15) can be obtained a variety of syntheses. The preferred routes are depicted in Schemes 7 and 8, respectively. It will be apparent to those skilled in the art that the following are representative examples, and that modifications of the disclosed synthetic protocols allow for the preparation of isoxazoline- and isoxazolinone intermediates.

Scheme 7.



As shown in Scheme 7, p-fluorobenzaldehyde (27) can be reacted with commercially available BOC-protected piperazine, 1-(t-butoxycarbonyl)piperazine (Aldrich Chemical Company, Inc., Milwaukee, WI), in the presence of potassium carbonate and a suitable solvent, such as pyridine (Py), and at a suitable temperature

PATENT APPLICATION

28341/6304.NDV1

(e.g., reflux) to provide piperazinyl intermediate (28). The formation of ester alcohol (29) results from the condensation of Compound (28) with ethyl diazoacetate, as described in Mahmood et al., *J. Org. Chem.*, 63, pgs. 3333-3336 (1998). Addition of hydroxylamine, followed by warming to reflux in aqueous methanol, yields 5 piperazinylarylisoxazolinone (30). Compound (30) then is converted to the corresponding methylacetamide (31) by reaction with N-(hydroxymethyl)acetamide acetate/DMF. The BOC group then is removed by treatment with an acid, preferably trifluoroacetic acid in dichloromethane (DCM), to yield piperazinylarylisoxazolinone Compound (12).

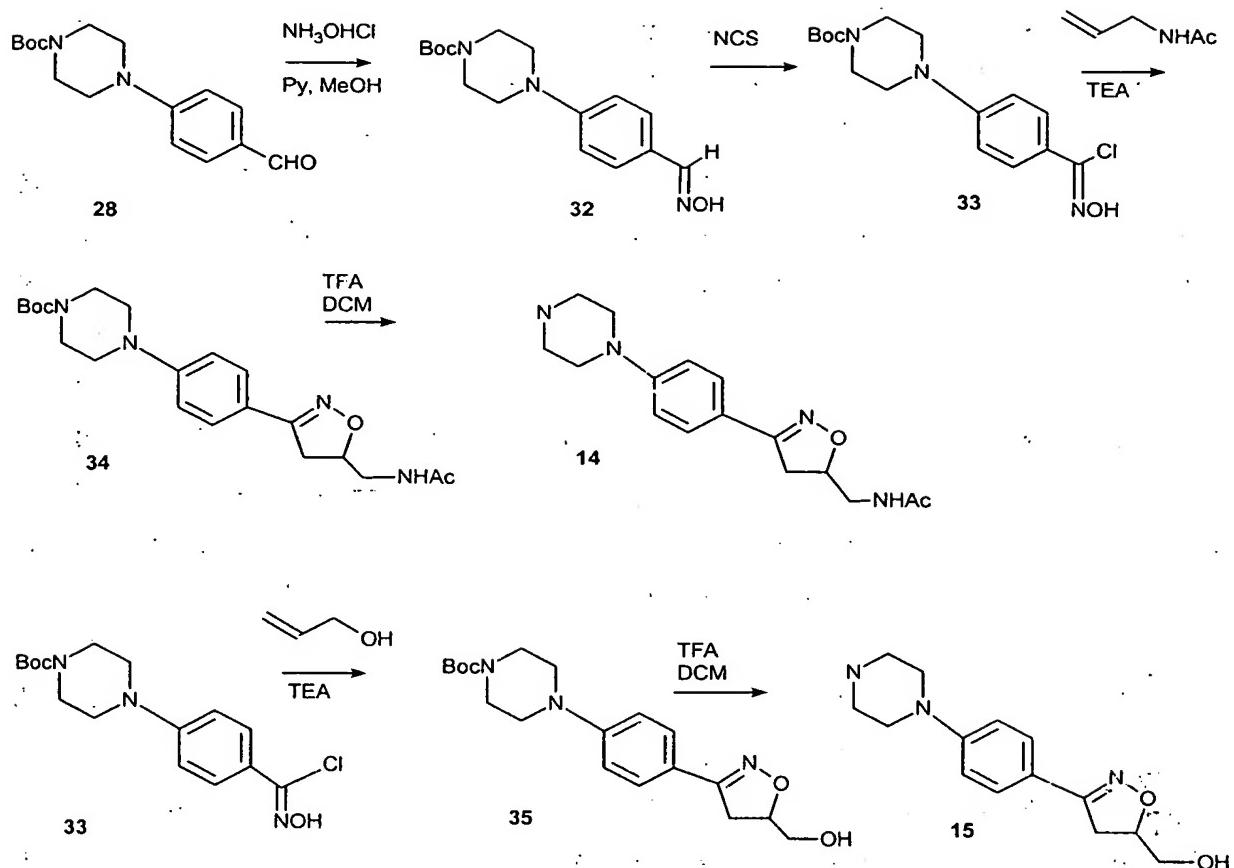
10.

As shown in Scheme 8, intermediate (28) also can be reacted with hydroxylamine hydrochloride in a polar protic solvent, such as methanol (MeOH), in the presence of a base, such as pyridine, to afford oxime (32). The oxime (32) then is oxidized by N-chlorosuccinamide (NCS) in an appropriate solvent, such as dichloromethane, to give the oximyl chloride (33). Alternatively, oximyl chloride (33) can be formed in situ and directly treated with an allylic compound such as N-acetylallylamine or allyl alcohol, in the presence of a suitable solvent, such as dichloromethane (DCM), to give compounds (34) and (35), respectively. The BOC protecting group then is removed by reaction with an acid, preferably trifluoroacetic acid in dichloromethane, to furnish piperazinylarylisoxazolinone compounds (14) and (15), respectively.

15

20

PATENT APPLICATION
28341/6304.NDV1
Scheme 8.



Other combinations of a quinolone with an oxazolidinone, an isoxazolinone, or an isoxazoline, also are possible. One embodiment involves covalent binding of the oxazolidinone, isoxazolinone, or isoxazoline to the side chains of the preferred C-7 quinolone substituents, for example the side chain amino group of optically active (amino)cycloalkylamino C-7 substituents.

The oxazolidinone- and isoxazoline-substituted quinolones (I) of the present invention contain at least one chiral center. It is apparent to one skilled in the art that when one chiral center is present, the compound can exist as one of two possible optical isomers ((R) and (S) enantiomers) or a racemic mixture of both. Both individual (R) and (S) enantiomers, as well as mixtures thereof, are within the scope of the oxazolidinone- and isoxazoline- substituted quinolones (I) of the invention. In the

PATENT APPLICATION

28341/6304.NDV1

event a second chiral center is present in the oxazolidinone- and isoxazoline-substituted quinolones (I) of the invention, the resultant diastereomers, in racemic and enantiomerically enriched forms, also are within the scope of the compounds (I) of the invention.

5

The preferred compounds of the present invention are optically pure enantiomers having the (S)-configuration at C⁵ of the oxazolidinone or isoxazoline ring, because, for example, S-ofloxacin exhibits a 10 to 100-fold greater potency than R-ofloxacin. However, the racemic mixture also is useful, but a greater amount of the 10 racemic material may be required to produce the same effect as the pure S-enantiomer.

If desired, the mixture of enantiomers is resolved by means known to those skilled in the art. Optically pure material can be obtained by resolution of the 15 racemic mixture by HPLC using a chiral phase, such as a Chiralpack AD column as described in Examples 4 and 6 for compounds 15 and 17 and shown in Scheme 2. Alternatively, resolution of the racemic mixture can be accomplished by selective crystallization of a salt form using methods known to those skilled in the art. See for example, "Optical Remixture Procedures for Chemical Compounds, Vol 1; Amines

20

and Related Compounds," Paul Newman, Optical Remixture Information Center, Manhattan College, Riverdale, N.Y., 10471, 1978. For example, treatment of the R,S-aminomethyl mixture (25) with an appropriate optically active acid, such as (+)-tartaric acid, or alternatively with (-)-tartaric acid, yields a mixture of diastereomeric salts, which can be separated by fractional crystallization to give a salt containing one 25 enantiomer of the racemic mixture. Other suitable optically active acids include (-)-dibenzoyl-tartaric acid, (+)-camphoric acid, (+)- and (-)-malic acid, and (+)-camphor-10-sulfonic acid. By reacting the diastereomeric salt with a base, the optically pure free amino compound (25) is obtained.

30

PATENT APPLICATION

28341/6304.NDV1

A compound of formula (I), or a prodrug or a physiologically acceptable salt or solvate thereof, can be administered as the neat compound or as a pharmaceutical composition containing either entity.

5 The pharmaceutical compositions of the present invention can be prepared by admixing a compound of formula (I) with a solid or liquid pharmaceutically acceptable carrier, and, optionally, with pharmaceutically acceptable adjuvants and excipients employing standard and conventional techniques. Solid form compositions include powders, tablets, dispersible granules, capsules, cachets and suppositories. A solid carrier can be at least one substance which also can function as a diluent, flavoring agent, solubilizer, lubricant, suspending agent, binder, tablet disintegrating agent, and encapsulating agent. Inert solid carriers include magnesium carbonate, magnesium stearate, talc, sugar, lactose, pectin, dextrin, starch, gelatin, cellulosic materials, a low melting wax, cocoa butter, and the like. Liquid 10 form compositions include solutions, suspensions, and emulsions. For example, compounds of the present invention can be dissolved in water, water-propylene glycol, or water-polyethylene glycol, optionally containing suitable conventional coloring agents, flavoring agents, stabilizers and thickening agents. The 15 oxazolidinone-, isoxazoline-, and isoxazolinone- substituted quinolones (I) can be used alone, or in conjunction with other antibacterial agents and/or non-antibacterial 20 agents, as known to those skilled in the art.

Pharmaceutically acceptable refers to those properties and/or substances which are acceptable from a pharmacological or toxicological point of view and from a physical or chemical point of view regarding composition, formulation, stability, patient acceptance, and bioavailability. Pharmaceutically acceptable hydrate means hydrates useful for administering the compounds of this invention, and suitable hydrates include the compounds complexed with at least one water molecule.

30

Pharmaceutically acceptable salts means salts useful for administering compounds of the present invention. Suitable salts include acid addition salts when a

PATENT APPLICATION

28341/6304.NDV1

basic group is present, such as occurs with the preferred piperazinyl group. Acid addition salts include those made from mineral acids, for example, hydrochloric, hydrobromic, hydroiodic, sulfuric, phosphoric, and the like, organic sulfonic acids, e.g., methanesulfonic, 2-hydroxyethyl sulfonates, organic carboxylic acids, e.g., amino and carbohydrate acids, e.g., gluconic, galacturonic, acetates, propionates, lactates, maleates, malates, succinates, tartrates, citric acid, fumarates, and the like. These salts can be in a hydrated form.

Pharmaceutically acceptable prodrugs means prodrugs useful for administering the compounds of this invention. Suitable prodrugs include acid derivatives, for example, amides, esters; for example, methyl esters, ethyl esters, and the like. It also is appreciated by those skilled in the art that the appropriate N-oxides of the nitrogens of the oxazolidinone-, isoxazoline-, and isoxazolinone- substituted quinolones (I) are included within the scope of the invention. These prodrugs also can be in a hydrated form.

Compounds and pharmaceutical compositions suitable for use in the present invention include those wherein the active ingredient is administered in an effective amount to achieve its intended purpose. More specifically, a "therapeutically effective amount" means an amount effective to prevent development of, or to alleviate the existing symptoms of, the subject being treated. Determination of the effective amounts is well within the capability of those skilled in the art, especially in light of the detailed disclosure provided herein.

A "therapeutically effective dose" refers to that amount of the compound that results in achieving the desired effect. Toxicity and therapeutic efficacy of such compounds can be determined by standard pharmaceutical procedures in cell cultures or experimental animals, e.g., for determining the LD₅₀ (the dose lethal to 50% of the population) and the ED₅₀ (the dose lethal to 50% of the population). The dose ratio between toxic and therapeutic effects is the therapeutic index, which is expressed as the ratio between LD₅₀ and ED₅₀. Compounds which exhibit high therapeutic indices are preferred. The data obtained from such data can

PATENT APPLICATION

28341/6304.NDV1

be used in formulating a dosage range for use in humans. The dosage of such compounds preferably lies within a range of circulating concentrations that include the ED₅₀ with little or no toxicity. The dosage can vary within this range depending upon the dosage form employed, and the route of administration utilized.

5

Humans and other mammals, for example, cattle, horses, sheep, hogs, dogs, and cats, can be treated with the oxazolidinone-, isoxazoline-, and isoxazolinone- substituted quinolones (I) of the present invention. The quinolones (I) of the present invention can be administered in a manner and in dosage forms similar to those of the known anti-bacterial agents described above. In therapeutic use for treating, or combating, bacterial infections in humans and warm-blooded animals, the compounds of formula (I), or pharmaceutical compositions thereof, are administered by conventional techniques, such as orally in solid and liquid dosage forms and/or parenterally (IV, IM, SQ), at a unit dosage form to obtain and maintain a concentration, that is, an amount, or blood-level of active component in the animal undergoing treatment which is antibacterially effective or appropriate.

10

15

20

25

30

Generally, the amount of compound (I) in a pharmaceutical composition is about 0.5% to about 90% by weight. An antibacterially effective dosage of compound (I) is about 0.1 to about 100 mg/kg of body weight/day, more preferably about 3 to about 50 mg/kg of body weight/day. The quantity of the oxazolidinone-, isoxazoline-, and isoxazolinone- substituted quinolone compounds of formula (I) in the pharmaceutical composition, the exact unit dosage form thereof to be administered, the frequency of administration, and the route of administration will vary, and can be adjusted widely depending upon a number of factors known to those skilled in the art including the particular mode of administration, the particular compound being used, the potency of the particular compound, the desired concentration, the age, weight, sex, and general physical condition and requirements of the patient, the nature and severity of the bacterial infection being treated, and the like, as is well known to the physician treating infectious diseases. Also, it is to be understood that the initial dosage administered can be increased beyond the above upper level in order to rapidly achieve the desired blood-level or the initial dosage.

PATENT APPLICATION

28341/6304.NDV1

can be smaller than the optimum and the daily dosage can be progressively increased during the course of treatment depending on the particular situation. The usual pharmaceutical dosage forms appropriate for parenteral (mixture, suspension in oil) and oral (tablet, capsule, syrup, suspension, etc) administration are known to those skilled in the art.

Compounds of the present invention can be administered by any suitable route, for example by oral, topical, buccal, inhalation, sublingual, rectal, vaginal, transurethral, nasal, topical, percutaneous, i.e., transdermal, or parenteral (including intravenous, intramuscular, subcutaneous, and intracoronary) administration. Parenteral administration can be accomplished using a needle and syringe, or using a high pressure technique, like POWDERJECT™.

If the compounds or pharmaceutical compositions of the present invention are administered parenterally, i.e., by injection, for example, by intravenous injection or by other parenteral routes of administration, it generally is as a soluble salt (acid addition salt or base salt) of the compound according to formula (I) in a pharmaceutically acceptable amount dissolved in a pharmaceutically acceptable liquid carrier such as, for example, water-for-injection, and a buffer to provide a suitable buffered isotonic solution, for example, having a pH of about 3.5 to about 6.

Suitable buffering agents include, for example, trisodium orthophosphate, sodium bicarbonate, sodium citrate, N-methylglucamine, L(+)-lysine, and L(+)-arginine. A compound of formula (I) generally is dissolved in the carrier in an amount sufficient to provide a pharmaceutically acceptable injectable concentration in the range of about 1 to about 400 mg/ml of solution. The resulting liquid pharmaceutical composition is administered so as to obtain the above-mentioned antibacterially effective amount of dosage.

For human use, a compound of the formula (I) can be administered alone, but generally is administered in admixture with a pharmaceutical carrier selected with regard to the intended route of administration and standard

PATENT APPLICATION

28341/6304.NDV1

pharmaceutical practice. Pharmaceutical compositions for use in accordance with the present invention can be formulated in a conventional manner using one or more physiologically acceptable carriers comprising excipients and auxiliaries that facilitate processing of compounds of formula (I) into preparations which can be used pharmaceutically.

These pharmaceutical compositions can be manufactured in a conventional manner, e.g., by conventional mixing, dissolving, granulating, dragee-making, levigating, emulsifying, encapsulating, entrapping, or lyophilizing processes.

Proper formulation is dependent upon the route of administration chosen. When a therapeutically effective amount of a compound of the present invention is administered orally, the composition typically is in the form of a tablet, capsule, powder, solution, or elixir. When administered in tablet form, the composition can additionally contain a solid carrier, such as a gelatin or an adjuvant. The tablet, capsule, and powder contain about 5 to about 95% compound of the present invention, and preferably from about 25 to about 90% compound of the present invention. When administered in liquid form, a liquid carrier such as water, petroleum, or oils of animal or plant origin can be added. The liquid form of the composition can further contain physiological saline solution, dextrose or other

saccharide solutions, or glycols. When administered in liquid form, the composition contains about 0.5 to about 90% by weight of a compound of the present invention, and preferably about 1 to about 50% of a compound of the present invention.

When a therapeutically effective amount of a compound of the present invention is administered by intravenous, cutaneous, or subcutaneous injection, the composition is in the form of a pyrogen-free, parenterally acceptable aqueous solution. The preparation of such parenterally acceptable solutions, having due regard to pH, isotonicity, stability, and the like, is within the skill in the art. A preferred composition for intravenous, cutaneous, or subcutaneous injection typically contains, in addition to a compound of the present invention, and isotonic vehicle.

PATENT APPLICATION
28341/6304.NDV1

For oral administration, the compounds can be formulated readily by combining a compound of formula (I) with pharmaceutically acceptable carriers well known in the art. Such carriers enable the present compounds to be formulated as tablets, pills, dragees, capsules, liquids, gels, syrups, slurries, suspensions and the like, for oral ingestion by a patient to be treated. Pharmaceutical preparations for oral use can be obtained by adding a compound of formula (I) with a solid excipient, optionally grinding a resulting mixture, and processing the mixture of granules, after adding suitable auxiliaries, if desired, to obtain tablets or dragee cores. Suitable excipients include, for example, fillers and cellulose preparations. If desired, disintegrating agents can be added.

For administration by inhalation, compounds of the present invention can be delivered in the form of an aerosol spray presentation from pressurized packs or a nebulizer, with the use of a suitable propellant. In the case of a pressurized aerosol, the dosage unit can be determined by providing a valve to deliver a metered amount. Capsules and cartridges of, e.g., gelatin, for use in an inhaler or insufflator can be formulated containing a powder mix of the compound and a suitable powder base such as lactose or starch.

The compounds can be formulated for parenteral administration by injection, e.g., by bolus injection or continuous infusion. Formulations for injection can be presented in unit dosage form, e.g., in ampules or in multidose containers, with an added preservative. The compositions can take such forms as suspensions, solutions, or emulsions in oily or aqueous vehicles, and can contain formulatory agents such as suspending, stabilizing, and/or dispersing agents.

Pharmaceutical formulations for parenteral administration include aqueous solutions of the active compounds in water-soluble form. Additionally, suspensions of the active compounds can be prepared as appropriate oily injection suspensions. Suitable lipophilic solvents or vehicles include fatty oils or synthetic fatty acid esters. Aqueous injection suspensions can contain substances which increase the viscosity of the suspension. Optionally, the suspension also can contain

PATENT APPLICATION

28341/6304.NDV1

suitable stabilizers or agents that increase the solubility of the compounds and allow for the preparation of highly concentrated solutions. Alternatively, a present composition can be in powder form for constitution with a suitable vehicle, e.g., sterile pyrogen-free water, before use.

5

Compounds of the present invention also can be formulated in rectal compositions, such as suppositories or retention enemas, e.g., containing conventional suppository bases. In addition to the formulations described previously, the compounds also can be formulated as a depot preparation. Such long-acting formulations can be administered by implantation (for example, subcutaneously or intramuscularly) or by intramuscular injection. Thus, for example, the compounds can be formulated with suitable polymeric or hydrophobic materials (for example, as an emulsion in an acceptable oil) or ion exchange resins, or as sparingly soluble derivatives, for example, as a sparingly soluble salt.

10

For topical administration, the present compounds can be applied in neat form, e.g., when the compound is a liquid. However, it is desirable to administer the compounds to the skin as compositions in combination with a dermatologically acceptable carrier, which can be a solid, semi-solid, or a liquid. Useful solid carriers include, but are not limited to, finely divided solids such as talc, clay, microcrystalline cellulose, silica, alumina, and the like. Useful liquid carriers include, but are not limited to, water, alcohols, glycols, and water-alcohol/glycol blends in which the present compounds can be dissolved or dispersed at effective levels, optionally with the aid of a surfactant. Adjuvants, such as fragrances and additional antimicrobial agents, can be added to optimize the properties for a given use. The resultant liquid compositions can be applied topically by absorbent pads, used to impregnate bandages and other dressings, or sprayed onto the affected area using pump-type or aerosol sprayers.

15

For veterinary use, a compound of formula (I) or a nontoxic salt thereof, is administered as a suitably acceptable formulation in accordance with

PATENT APPLICATION

28341/6304.NDV1

normal veterinary practice. The veterinarian can readily determine the dosing regimen and route of administration that is most appropriate for a particular animal.

General Methods and Definitions

5

Reagents were purchased from commercial sources and used without further purification. All temperatures are in degrees Centigrade. When solvent pairs are used, the ratios of solvents used are volume/volume (v/v). When the solubility of a solid in a solvent is used the ratio of the solid to the solvent is weight/volume (wt/v).

10 Reactions with moisture-sensitive reagents were performed under a nitrogen atmosphere. Concentration of solutions was performed by reduced pressure rotary evaporation. Brine refers to an aqueous saturated sodium chloride mixture. High performance liquid chromatography (HPLC) analysis and purification were performed using Beckman System Gold®; detection at 220 nm. Analytical HPLC was performed
15 on a YMC 5 micron C18 (4.6 mm x 50 mm) reverse phase (RP) column (gradient from 100% of the aq. 0.1% trifluoroacetic acid (TFA) to 100% of 0.1% TFA in acetonitrile (MeCN) over 6 min, flow rate 2.0 mL/min). Preparative thin-layer chromatography (TLC) were performed using EM silica gel (SG) 60 F₂₅₄ plates (20 x 20 cm, thickness 2 mm). NMR refers to nuclear magnetic resonance spectroscopy.
20 ¹H NMR refers to proton nuclear magnetic resonance spectroscopy with chemical shifts reported in ppm downfield from tetramethylsilane. Mass-spectra (MS) refers to mass spectrometry expressed as m/e or mass/charge unit and was obtained using electron impact (EI) technique. [M+H]⁺ refers to the positive ion of a parent plus a hydrogen atom. Retention time (R_t) is in minutes and refers to x. IR refers to infrared
25 spectroscopy. FTIR refers to Fourier Transform IR.

In the following examples, the following abbreviations are used:
millimole (mmol), milliliter (mL), potassium carbonate (K₂CO₃), ethyl acetate (EtOAc), DMSO (dimethyl sulfoxide), magnesium sulfate (MgSO₄), sodium
30 bicarbonate (NaHCO₃), and ethyl alcohol (EtOH).

EXAMPLES

5 The following examples describe how to prepare the various compounds and/or perform the various processes of the invention, and are to be construed as merely illustrative, and not limitations of the preceding disclosure in any way whatsoever. Those skilled in the art will recognize appropriate variations from the procedures both as to reagents and as to reaction conditions and techniques.

10

General Procedure for the Synthesis of Quinolone-7-yl Linked Quinolone-Oxazolidinones and Related Analogs.

A mixture of an appropriate nucleophile linked oxazolidinone, or an oxazolidinone precursor such as an amino alcohol, (1-2 mmol) with an appropriate 7-substituted quinolone (preferably, 7-fluoro, 7-chloro, or 7-triflate derivative) (1 mmol) in N-methylpyrrolidine-2-one (NMP) (2 mL), N-methylmorpholine (NMM) (0.4 mL) and (optionally, DMSO, as an additional co-solvent, is stirred at 110-130 °C for 24-96 h (preferably, 24-48 h (hours) and 96 h for reactions with 7-fluoro and 7-chloroquinolones, respectively). The mixture is cooled to room temperature (r.t.), and a majority of the solvent removed under vacuum. The residue is triturated with water (7 mL), precipitated, filtered, washed with excess water, THF (ca. 4 x 15 mL), ether, and dried under vacuum. Optionally, the resulting intermediate amino alcohol derivative is purified by crystallization from an appropriate solvent (e.g., MeOH) or by silica gel column chromatography (eluent: dichloromethane-MeOH). The amino alcohol (1 mmol) is dissolved in a aprotic organic solvent (e.g., tetrahydrofuran (THF) or NMP) (2-5 mL), and carbonyldiimidazole (1.1 mmol) is added. Optionally, an organic base is added (e.g., imidazole) (1.1 mmol). The mixture is stirred at 20 - 40 °C for 1 - 3 h. The solvent is removed under vacuum, and the crude product is purified by crystallization from an appropriate solvent (e.g., MeOH) or by silica gel column chromatography (eluent: dichloromethane-MeOH). Optionally, the carboxylic functionality is esterified under standard alcohol coupling conditions (e.g.,

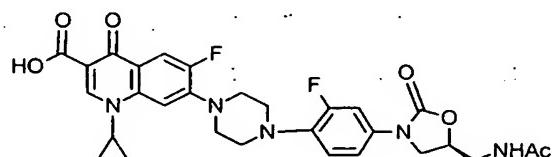
PATENT APPLICATION

28341/6304.NDV1

polyethyleneglycol with diethyl cyanophosphate, 4-dimethylaminopyridine in NMP at 20-40 °C for 4-8 h) to provide an ester prodrug derivative.

EXAMPLE 1

5



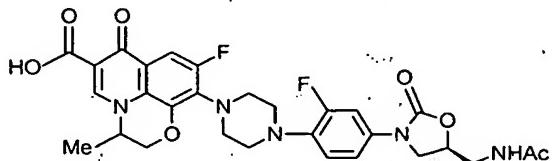
Preparation of

10 **7-[4-[(S)-Acetamidomethyl]oxazolidine-2-one-3-yl]-2-fluorophenylpiperazine-1-yl]-3-carboxy-1-cyclopropyl-6-fluoro-1,4-dihydroquinoline-4-one (Compound 3).**

15 Compound 3 was prepared from 5-(S)-aminomethyl-3-(3-fluoro-4-piperazinophenyl)oxazolidine-2-one (0.372 g, 1.1 mmol) and 3-carboxy-1-cyclopropyl-7-chloro-6-fluoro-1,4-dihydroquinoline-4-one (0.282 g, 1 mmol) according to the General Procedure for the Synthesis of Quinolone-7-yl Linked Oxazolidinones. The reaction was performed at 120 °C for 96 h. Yield 0.36 g (62%). MS(m/z): 582 [M+H]⁺. R_t 4.6 min.

20

EXAMPLE 2

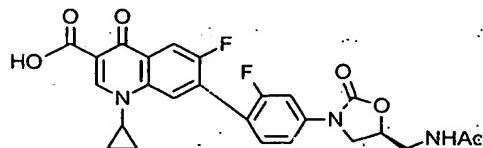


PATENT APPLICATION
28341/6304.NDV1

Preparation of 10-[4-[(5-(S)-Acetamidomethyl)oxazolidine-2-one-3-yl]-2-fluorophenyl]piperazine -1-yl]-6-carboxy-9-fluoro-2,3-dihydro-3-methyl-7-oxo-7H-pyrido[1,2,3-de]-1,4-benzoxazine (Compound 6).

5 Compound 6 was prepared from 5-(S)-aminomethyl-3-(3-fluoro-4-piperazinophenyl)oxazolidine-2-one (0.372 g, 1.1 mmol) and 9,10-difluoro-2,3-dihydro-3-methyl-7-oxo-7H-pyrido[1,2,3-de]-1,4-benzoxazine-6-carboxylic acid (0.281 g, 1 mmol) according to the General Procedure for the Synthesis of Quinolone-7-yl Linked Oxazolidinones. The reaction was performed at 10 110 °C for 24 h. Yield 0.444 g (74%). MS (m/z): 598 [M+H]⁺. R_t 4.5 min. The separation of enantiomers by chiral HPLC provides the preferred (S)-configuration isomer.

EXAMPLE 3



15 **Preparation of 7-(4{(5S)-5-[(Acetylamino)methyl]2-oxo-1,3-oxazolidin-3-yl}-2-fluorophenyl)-1-cyclopropyl-6-fluoro-4-oxo-1,4-dihydro-3-quinolinecarboxylic acid (Compound 11).**

20 **Benzyl-1-cyclopropyl-6-fluoro-4-oxo-7-[(trifluoromethyl)sulfonyl]oxy-1,4-dihydro-3-quinolinecarboxylate (Compound 7a).**

A slurry of 1-cyclopropyl-6-fluoro-4-oxo-7-hydroxy-1,4-dihydro-3-quinolinecarboxylate (1.51 g, 5.75 moles), prepared as described in Kiely, J.S.; Laborde, E.; Lesheshki, L.E.; Busch, R.A. *J. Heterocyclic Chem.* 1991, 28, 1581-1585, in dry pyridine (15 mL) was cooled to 0°C and treated via syringe with

PATENT APPLICATION

28341/6304.NDV1

trifluoromethanesulfonic anhydride (2.5 mL, 14.86 moles). The resulting homogeneous, amber-colored solution was warmed slowly to room temperature and stirred for 24 hours. Benzyl alcohol (20 mL, 193.3 moles) was added, and the reaction stirred at room temperature for 2 hours. After pouring the solution into 100 mL of water, the aqueous phase was extracted (3X) with CH₂Cl₂. The combined organic layers were dried (Na₂SO₄), filtered and evaporated. The crude product was chromatographed on a Biotage 40 g column. The column was conditioned and loaded with CH₂Cl₂ and eluted with 850 mL of CH₂Cl₂ and 800 mL of 5% MeOH/CH₂Cl₂. Fractions 7-13 (A) and fractions 27-28 (B) (~45 mL cuts) were combined and evaporated, but neither was pure. Impure fraction A contained benzyl alcohol, which was removed in a nitrogen stream. The resulting solids were triturated with EtOAc, filtered and dried (House vacuum, 55EC, 1 hour) to yield Compound 7a as a white solid (36A) that weighed 99.0 mg (3.5%). Another 67 mg of 7a was obtained from the filtrate. Impure fraction B was recrystallized from MeOH/CH₂Cl₂ and EtOAc. The solids (36B) were collected by suction filtration and dried (house vacuum, 55EC, 1 hour) to afford Compound 7a as an off-white solid that weighed 285 mg (10%). Another 235 mg of Compound 7a was obtained in the mother liquors. ES-MS of these products did not provide useful information. ¹H NMR (DMS)-d₆, TMS): * 8.54 (s, 1 H), 8.30 (d, J = 8 Hz, 1 H), 8.04 (d, J = 12 Hz, 1 H), 7.49 (m, 2 H), 7.42-7.33 (m, 3 H), 5.29 (s, 2 H), 3.69 (m, 1 H), 1.26 (m, 2 H), 1.11 (m, 2 H). The ¹H NMR of 36B was identical to that of 36A. TLC (of 36A and 36B): R_f=0.67 (5% EtOAc/MeOH); UV--visualization

Preparation of *tert*-Butyl [(5*S*)-3-(4-borono-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl] methylcarbamate (Compound 8)

2-Methylpropyl (4-bromo-3-fluorophenyl)carbamate.

To a solution of 500 g (4.50 mol) of 3-fluoroaniline and 2 L of CH₂Cl₂ in a 12 L round bottom flask was added a solution of 473 g (3.42 mol, 0.76 equiv.) of K₂CO₃ in 2 L of water. Isobutylchloroformate (663 g, 4.86 mol, 1.08 equiv.) was

PATENT APPLICATION

28341/6304.NDV1

added with stirring via an addition funnel over 3 h so as to allow the isotherm to warm and maintain the mixture at gentle reflux. Gas evolves vigorously near the end of the addition. The organic phase was sampled for GC analysis 1 h after completion of the addition; less than 0.5% 3-fluoroaniline remained. The mixture was quenched by 5 addition of 72 mL of conc. NH₄OH. After stirring for 15 minutes, the mixture was neutralized to pH by addition of 120 mL of conc. aq. HCl. The layers were separated, and the aqueous layer was extracted with 1.5 L of CH₂Cl₂. To the combined organic layers was added 987 g (3.45 mol, 0.77 equiv.) of 1,3-dibromo-5,5-dimethylhydantoin and 2.5 L of water. The mixture was allowed to isotherm to 39E C and held at that 10 temperature by gentle heating for 2 hours. The reaction was judged complete by HPLC analysis of the organic layer. The mixture was cooled to 32E C by addition of 500 g of ice, and the solids not soluble in either liquid layer (mostly hydantoins and partially brominated hydantoins) were removed by filtration. The filtrate layers were separated, extracting the aqueous layer with 500 mL of CH₂Cl₂. The combined 15 organic layers were added with rapid stirring to a solution of 410 g of Na₂SO₃ in 3 L of water. The layers were separated, extracting the aqueous layer with 3400 mL of CH₂Cl₂. The combined organics were distilled on a rotovap and replaced with heptane, maintaining a constant volume. the resulting thick slurry was cooled to 4E over 2.5 h. The solids were collected by filtration, washed with heptane (2 x 750 mL) and air dried, affording 1097g (84%) of 2-Methylpropyl (4-bromo-3-fluorophenyl)carbamate as a white crystalline solid.

(5R)-3-(4-bromo--3-fluorophenyl)-5-(hydroxymethyl)-1,3-oxazolidin-2-one.

25 To a solution of 1056 g (3.64 mol) of 2-Methylpropyl (4-bromo-3-fluorophenyl)carbamate in 6.65 L of THF cooled to -15E C in a 22 L round bottom flask was added a solution of 428g (4.55 mol, 1.25 equiv.) of lithium t-amylate over 10 min via an addition funnel, maintaining -15E to -12E C. In a separate 5 L flask, a solution of 438g (4.37 mol, 1.20 equiv.) of (S)-3-chloro-1,2-propanediol in 1.75 L of THF was cooled to -25 E C and treated with a 20% t-BuOK solution in THF (2645 mL, 4.29 mol, 1.18 equiv) over 25 min, resulting in a thick but stirrable slurry. This

PATENT APPLICATION

28341/6304.NDV1

was allowed to warm to 10E over 75 min and then poured into the 22 L flask containing the carbamate solution. The resulting slurry was allowed to warm from -7E to 7E over 1.5 h, monitoring reaction progress by HPLC. Upon completion (~2.5% each of remaining 2-Methylpropyl (4-bromo-3-fluorophenyl)carbamate and the over addition product) a quench solution composed of 1.05 AcOH and 3.5 L of water was added. The layers were separated. The aqueous layer was back-extracted with 1 L of THF, and the combined organic layers were washed with brine. The volatile were removed, giving white solids wet with acetic acid. This material was slurried in 1.6 L

10 of EtOAc. Hexane (4L) was added over 1 h. The resulting slurry was cooled to 2E over 1 h and filtered, giving 916 g (87%) of (5R)-3-(4-bromo-3-fluorophenyl)-5-(hydroxymethyl)-1,3-oxazolidin-2-one as coarse white crystals: TLC $R_f = 0.008$ (50% EtOAc/hexane); HPLC rt = 2.55 min; mp 114--121E; $[\nabla]_D = +52.2E$ c = 1, MeOH); 1H NMR (DMSO) * 7.49 (m, 2H), 7.15 (d, 1H, J = 8.5 Hz), 5.30 (br s, 1H), 4.54 (m, 1H), 3.89 (t, 1H, J = 8.6 Hz), 3.67 (m, 1H), 3.50 (dd, 1H, J = 3.0, 11.8 Hz), 3.39 (dd, 1H, 3.8, 11.8 Hz); ^{13}C NMR (DMSO) * 158.1 (s, $J_{CF} = 241$ hz), 154.2 (S), 139.7 (S, $J_{CF} = 10$ hz), 133.3 (D), 114.9 (D), 105.9 (d, $J_{CF} = 29$ Hz), 100.9 (s, $J_{CF} = 21$ Hz); 73.3(d), 61.5(t), 45.9 (t); Anal. calc'd for $C_{10}H_9BrFNO_3$: C, 41.40; H, 3.13; N, 4.83; Br, 27.55; found: C, 41.11; H, 3.06; N, 4.83; Br, 26.97.

20
[(5R)-3-(4-bromo-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl]methyl 3-nitrobenzenesulfonate.

To a slurry of 907 g (3.13 mol) of (5R)-3-(4-bromo-3-fluorophenyl)-5-(hydroxymethyl)-1,3-oxazolidin-2-one in 4.5 L of CH_2Cl_2 in a 22 L round bottom flask was added 654 mL (4.69 mol, 1.50 equiv.) of triethylamine. The mixture was cooled to 0E C, and a solution of 832 g (34.75 mol, 1.20 equiv) of m-nitrobenzenesulfonyl chloride in 2 L of CH_2Cl_2 was added, keeping the temperature below 6E C. The mixture was sampled for HPLC, and an additional 55 g of solid m-nitrobenzenesulfonyl chloride was added to drive the reaction to completion. Hydrochloric acid (1M, 4.5 L) was added to the slurry. The solids were collected by

PATENT APPLICATION

28341/6304.NDV1

filtration and washed with water. The phases in the filtrate were separated, and the aqueous later was extracted with 2 x 500mL of CH₂Cl₂. The combined organic layers were concentrated and combined with the solids previously isolated, 2 L of CH₂Cl₂ and 2 L of methanol. This slurry was distilled on a rotary evaporator, keeping the volume at ~5 L by adding methanol. The solids were collected by filtration, washed with 1 L of MeOH, and air dried to afford 1415 g (95%) of [(5R)-3-(4-bromo-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl] methyl 3-nitrobenzene sulfonate: TLC R_f = 0.15 (50% EtOAc/hexanes); HPLC rt = 5.68 min; mp 153-156E; [Δ]_D = -76.6E C = 1, MeOH/CH₂Cl₂; ¹H NMR (DMSO) *8.77 (d, 1H, J = 8.2 Hz), 8.71 (s, 1H), 8.52 (d, 1H, 7.9 Hz), 8.14 (t, 1H, J = 8.1 Hz), 7.85 (t, 1H, J = 8.5 Hz), 7.73 (dd, 1H, J=2.3, 11.4 Hz), 7.40 (d, 1H, J = 8.9 Hz), 5.12 (m, 1H), 4.68 (m, 2H), 4.28 (t, 1H, J = 9.4Hz), 3.89 (m, 1H); ¹³C NMR (DMSO) * 157.3 (s, J_{CF} = 242 hz), 152.4 (S), 147.2 (S), 138.3 (S, J_{CF} = 1.0 hz), 135.4 (S), 132.7 (D), 132.6 (D), 131.1 (D), 128.2 (D), 121.7 (D), 114.2 (D), 105.3 (D, J_{CF} = 29 Hz), 100.6 (s, J_{CF} = 20 Hz), 70.4 (t), 69.0 (d), 44.8 (t); Anal. calc'd for C₁₆H₁₂BrFN₂O₇S: C, 40.44; H, 2.55; N, 5.89; Br, 16.81; found: C, 40.22; H, 2.45; N, 5.86; Br, 16.60.

tert-Butyl [(5*S*)-3-(4-bromo-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl] methylcarbamate.

In three portions, the sulfonate [(5*R*)-3-(4-bromo-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl] methyl 3-nitrobenzene sulfonate(1400 g., 2.95 mol) was suspended in 15mL/g of a solvent mixture of 29% aqueous NH₄OH, MeCN, and MeOH in a 5:2.5:1 ratio in an autoclave. The system was sealed and heated to 80E C for 3-4 h with stirring. Upon cooling, the mixtures were extracted three times each with CH₂Cl₂. the combined extracts were concentrated, giving the crude amine as solid. The solids were suspended in 8.5 L of CH₂Cl₂. Di-*t*-butyldicarbonate (985 g, 4.42 mol, 1.5 equiv.) was added as a solid over 15 min, with vigorous gas evolution. The mixture was stirred at room temperature overnight, after which the reaction was complete by TLC. Water (3L) was added, and stirring was continued for 30 min. The mixture was filtered, washing the solids that collected with additional CH₂Cl₂. The

PATENT APPLICATION

28341/6304.NDV1

layers in the filtrate were separated. The organic layer was concentrated to a white solid, crude *tert*-butyl [(5*S*)-3-(4-bromo-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl] methylcarbamate. This was suspended in 3L of EtOAc and heated to 70 EC to dissolve. Upon cooling to room temperature, 3L of hexane was added over 1h. The resulting slurry was cooled to 0E C. The solids were collected by filtration and washed with hexane to obtain 763 g of *tert*-butyl [(5*S*)-3-(4-bromo-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl] methylcarbamate. A second crop was obtained by concentrating the mother liquor to 1.2 L and cooling to 0E C, yielding an additional 50 g (total 813 g, 71%): TLC R_f = 0.31 (50% EtOAc/hexane); HPLC rt = 5.02 min; mp 145-146E; [V]_D = +30.0E c = 1, MeOH); ¹H NMR (CDCl₃) * 7.50 (m, 2H), 7.12 (d, 1H, 8.5 Hz), 5.04 (br s, 1H), 4.77 (m, 1H), 4.01 (t, 1H, J = 8.7 Hz), 3.84 (m, 1H), 3.53 (m, 2H), 1.40 (S, 9H); ¹³C NMR (CDCl₃) * 160.2(s), 157.3 (s, J_{CF} = 242 Hz), 154.0 (S), 138.8 (s, J_{CF} = 9 Hz), 133.5 (d), 114.4 (d), 106.8 (d, J_{CF} = 28 Hz), 103.3 (s, J_{CF} = 21 Hz), 80.4 (s), 72.1 (d), 47.3 (t), 43.2 (t), 28.2 (q); Anal. calc'd for C₁₅H₁₈BrFN₂O₄: C, 46.29; H, 4.66; N, 7.20; Br, 20.53; found: C, 46.32, H, 4.57; N, 7.21, Br, 20.63.

***tert*-Butyl [(5*S*)-3-(4-borono-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl] methylcarbamate (Compound 8).**

To a solution of 2.00g (5.14 mmol) of *tert*-Butyl [(5*S*)-3-(4-bromo-3-fluorophenyl)-2-oxo-1,3-oxazolidin-5-yl] methylcarbamate in 40 mL of THF was added 1.94 mL (1.49 g, 12.8 mmol, 2.50 equiv.) of N,N,N',N'-tetramethylenediamine. The solution was cooled to -50E, and 4.86 mL (5.45 mmol, 1.06 equiv.) of 1.12 M ethyl magnesium bromide in THF solution was added by syringe. After 5 min., 4.06 mL (10.8 mmol, 2.10 equiv.) of 2.66 M n-butyl lithium in hexane solution was added dropwise by syringe with vigorous stirring, keeping the temperature below 45 EC. A thick slurry resulted, which was stirred for an additional 15 min. Trimethylborate (1.17 mL, 1.07 g, 10.3 mmol, 2.00 equiv.) was added by syringe. The mixture was allowed to warm to 20 EC over 90 min. It was then poured into 25 mL of 4M hydrochloric acid and stirred for 15 min. The layers were separated, and the aqueous layer was extracted with 20 mL of CH₂Cl₂ to complete the removal

PATENT APPLICATION

28341/6304.NDV1

of boronic acid. The combined organic extracts were dried ($MgSO_4$), filtered, and concentrated to give crude Compound 8 as an oil.

5 Benzyl-7-[4-((5S)-{[(tert-butoxycarbonyl)amino]methyl}-2-oxo-1,3-oxazolidin-3-yl)-2-fluorophenyl]-1-cyclopropyl-6-fluoro-4-oxo-1,4-dihydro-3-quinolinecarboxylate (Compound 9a).

A mixture of Compound 7a (369 mg, 0.76 moles) and Compound 8 (298 mg, 0.84 moles) in 1,2 dimethoxythane (8 mL) was degassed and flushed with nitrogen several times. Dichlorobis(triphenylphosphine)palladium(II) (56.3 mg, 0.08 moles) and 2M K_2HPO_4 (0.77 mL, 1.54 moles) was added. The mixture was degassed and flushed with nitrogen several times and then heated at 90EC for 22 hours. After cooling to room temperature, the reaction was concentrated under reduced pressure, and the residue was chromatographed on a Biotage 40 gram column. The column was conditioned with 3:3:4 $CH_2Cl_2/EtOAc/heptane$, loaded with CH_2Cl_2 and eluted with 900 mL of 3:3:4 $CH_2Cl_2/EtOAc/heptane$ and 500 mL of EtOAc. Fractions 23-49 (~20 mL cuts) were combined, evaporated and dried (high vacuum, r.t., 1 hour) to yield 257.1 mg (52%) of Compound 9a as a tan, amorphous solid. MS (ESI+) for $C_{35}H_{33}F_2N_3O_7$ m/z 646.3 ($M+H$)⁺. ¹H NMR ($CDCl_3$, TMS); δ 8.64 (S, 1 H), 8.24 (D, $J = 13$ Hz, 1 H), 7.98 (D, $J = 7$ Hz, 1 H), 7.71-7.61 (m, 2 H), 7.55-7.47 (m, 3 H), 7.42-7.33 (m, 3 H), 5.42 (s, 2 H), 5.03 (m, 1 H), 4.82 (m, 1 H), 4.11 (m, 1 H), 3.94 (m, 1 H), 3.57 (m, 2 H), 3.49 (m, 1 H), 1.43 (s, 9 H), 1.33 (m, 2 H), 1.16 (m, 2 H). TLC: $R_f = 0.14$ (80% EtOAc/hexane); UV-visualized.

25 Benzyl-7-(4-{(5S)-5-[(acetylamino)methyl]-2-oxo-1,3-oxazolidin-3-yl}-2-fluorophenyl)-1-cyclopropyl-6-fluoro-4-oxo-1,4-dihydro-3-quinolinecarboxylate (Compound 10a).

A solution of Compound 9a (253 mg, 0.391 moles) in 5.2 mL of CH_2Cl_2 at 0EC was treated via syringe with trifluoroacetic acid (2.6 mL, 33.7 moles). The resulting solution was stirred at 0EC for 30 minutes and at room temperature for

PATENT APPLICATION
28341/6304.NDV1

1.5 hours. After concentrating the reaction under reduced pressure, the residue was dried (high vacuum, r.t., 2 hours) to afford the trifluoroacetic acid salt as an amber, amorphous solid (372 mg, over theory). The above salt was dissolved in 6.5 mL of pyridine at room temperature and treated with acetic anhydride (1.5 mL, 15.9 moles).

5 After stirring at room temperature for 17 hours, the reaction was cooled to 0EC and quenched with 1.5 mL of MeOH. The reaction was stirred at 0EC for 10 minutes and at room temperature for 20 minutes. Solvent was removed under reduced pressure, and the crude product (863 mg) was chromatographed on a Biotage 40 gram column. The column was conditioned with EtOAc, loaded with EtOAc (plus a small amount of

10 CH_2Cl_2) and eluted with 500 mL of EtOAc, 500 mL of 2% MeOH/ CH_2Cl_2 and 500 mL of 5% MeOH/ CH_2Cl_2 . Slightly impure fractions 42-70 (~20 mL cuts) were combined, evaporated (271 mg) and chromatographed again on a Biotage 8 gram column. The column was conditioned, loaded and eluted with 5% MeOH/ CH_2Cl_2 . Fractions 7-44 (about 7 mL cuts) were combined, evaporated and dried (house

15 vacuum, r.t., 16 hours) to generate 218.5 mg (95%) of Compound **10a** as a beige solid. MS (ESI+) for $\text{C}_{32}\text{H}_{27}\text{F}_2\text{N}_3\text{O}_6$ m/z 588.2 ($\text{M}+\text{H}$)⁺. ¹H NMR (CDCl_3 , + 2 drops CD_3OD , TMS): * 8.70 (s, 1 H), 8.17 (d, $J = 10$ Hz, 1 H), 8.03 (m, 1 H), 7.60 (m, 1 H), 7.50-7.46 (m, 3 H), 7.41-7.31 (m, 4 H), 5.41 (s, 2 H), 4.83 (m, 1 H), 4.11 (t, $J = 9$ Hz, 1 H), 3.86 (m, 1 H), 3.66 (m, 2 H), 3.54 (m, 1 H), 2.03 (s, 3 H), 1.36 (m, 2 H), 1.18 (m, 2 H). TLC: $R_f = 0.28$ (5% MeOH/ CH_2Cl_2); UV-visualized

20

7-(4-{(5S)-5-[(Acetyl amino)methyl]-2-oxo-1,3-oxazolidin-3-yl}-2-fluorophenyl)-1-cyclopropyl-6-fluoro-4-oxo-1,4-dihydro-3-quinolinecarboxylic acid (Compound 11).

25 A solution of Compound **10a** (215 mg, 0.365 moles) in absolute ethanol was treated with 30 mg of 10% Pd/C. The mixture was placed on the hydrogenator at room temperature at 40 psi H_2 for 3 hours. Since TLC analysis indicated the presence of starting material, an additional 30 mg of catalyst was added, and the reaction was returned to the hydrogenator at 40 psi of H_2 for 16 hours. After removing the catalyst by filtration through a pad of Elite and washing the filter cake

30

PATENT APPLICATION

28341/6304.NDV1

with absolute ethanol, the filtrates were combined and concentrated leaving a yellowish-black residue (160 mg). The residue was chromatographed on a Biotage 8 g column. The column was conditioned, loaded and eluted with 5% MeOH/CH₂Cl₂, but only mixed fractions were obtained. Fractions containing the desired product were combined and concentrated leaving a solid that was crystallized from EtOAc/heptane. TLC analysis of the solids (22 mg) revealed that crystallization did not upgrade the product. Therefore, the solids and mother liquors were dissolved in a minimal amount of MeOH/CH₂Cl₂ and loaded onto two 500:m prep TLC plates. After eluting once with 5% MeOH/CH₂Cl₂, the plates were eluted a second time with 5% MeOH/CH₂Cl₂ + 0.5% HOAc. Separation was not perfect, but a clean sample of the desired band was isolated affording 21.8 mg (12%) of Compound 11 as a gold solid. This product decomposed at 120EC. MS (ESI+) for C₂₅H₂₁F₂N₃)₆ m/z 498.2 (M+H)⁺. High resolution MS (FAB): Cal'd for C₂₅H₂₁F₂N₃O₆+H, 498.1476; Found, 498.1474. ¹H NMR (CDCl₃ + 1 drop CD₃OD, TMS): * 8.890 (s, 1 H), 8.24 (d, J = 13 Hz, 1 H), 8.16 (d, J = 8 HZ, 1 H), 7.66 (m, 1 H), 7.52 (m, 1 H), 7.38 (m, 1 H), 4.86 (m, 1 H); 4.14 (m, 1 H), 3.89 (m, 1 H), 3.70-3.61 (m, 3 H), 2.05 (s, 3 H), 1.41 (m, 2 H), 1.25 (m, 2 H). TLC: R_f = 0.22 (5% MeOH/CH₂Cl₂ + 1% HOAc); UV-visualized).

Ethyl 7-[4-((5S{[(tert-butoxycarbonyl)amino]methyl}-2-oxo-1,3-oxazolidin-3-yl)-2-fluorophenyl]-1-cyclopropyl-6-fluoro-4-oxo-1,4-dihydro-3-quinolinecarboxylate (Compound 9b).

A suspension of Compound 7b (380 mg., 0.898 moles), prepared as described in Kiely, J.S.; Laborde, E.; Lesheshki, L.E.; Busch, R.A. *J. Heterocyclic Chem.* 1991, 28, 1581-1585, and Compound 8 (350 mg, 0.988 moles) in 9 mL of 1,2-dimethoxythane was degassed by repeated evacuation and flushing with nitrogen. addition of 2M K₂HPO₄ (0.9 mL, 1.8 moles) was made via pipette followed by addition of dichlorobis(triphenylphosphine)palladium(II) (65 mg, 0.093 moles). The resulting mixture was degassed and refluxed under nitrogen for 21 hours. The dark brown, homogeneous solution was cooled to room temperature and concentrated under reduced pressure. The crude product (685 mg) was chromatographed on a 40S

PATENT APPLICATION

28341/6304.NDV1

Biotage column. The column was conditioned with 80% EtOAc/heptane, loaded with EtOAc (plus a small amount of CH₂Cl₂) and eluted with 900 mL of 80% EtOAc/heptane followed by 500 mL of EtOAc. Fractions 30-52 (20 mL cuts) were combined, evaporated and dried (house vacuum, 45E C, 3 days) to yield Compound 5 9b (175.5 mg, 33.5%) as a cream-colored solid. MS (ESI+) for C₃₀H₃₁F₂N₃O₇: m/z 584.1 (M+H)⁺. ¹H NMR (CDCl₃, TMS): * 8.65 (s, 1 H); 8.23 (d, J = 10.0 Hz, 1 H), 7.99 (d, J = 5.8 Hz, 1 H), 7.63 (m, 1 H), 7.50 (m, 1 H), 7.36 (m, 1 H), 5.01 (m, 1 H), 4.82 (m, 1 H), 4.42 (q, J = 7.1 Hz, 2 H), 4.10 (M, 1 H); 3.94 (m, 1 H); 3.58-3.50 (m, 3 H), 1.45-1.42 (m, 12 H), 1.34 (m, 2 H), 1.18 (m, 2 H). TLC: R_f = 0.32 (5% 10 MeOH/CH₂Cl₂); UV-visualization.

Ethyl 7-(4-{(5S)-5-[(acetylamino)methyl]-2-oxo-1,3-oxazolidin-3-yl}-2-fluorophenyl)-1-cyclopropyl-6-fluoro-4-oxo-1,4-dihydro-3-quinolinecarboxylate (Compound 10b).

A solution of Compound 9b (90 mg, 0.154 moles) in 2 mL of 15 methylene chloride at 0EC was treated via syringe with trifluoroacetic acid (1.0 mL, 12.98 moles). The resulting yellow solution was stirred at 0EC for 30 minutes and at room temperature for 1 hour. TLC analysis indicated that the reaction was complete. 20 The solvent was removed under reduced pressure, and the residue was dried under high vacuum at room temperature for 2 hours to afford ethyl-7{40[(5S)-5-(aminomethyl)-2-oxo-1,3-oxazolidin-3-yl]-2-fluorophenyl}-1-cyclopropyl-6-fluoro-4-oxo-1,4-dihydro-3-quinolinecarboxylate, trifluoroacetic acid salt (94.9 mg, quantitative) as a sticky, gold solid. MS:(ESI+) for C₂₅H₂₃F₂N₃O₅: m/z 484.4 25 (M+H)⁺. TLC: R_f = 0.19 (4% MeOH/CH₂Cl₂ + 1% NH₄OH); UV-visualization. A suspension of ethyl-7{40[(5S)-5-(aminomethyl)-2-oxo-1,3-oxazolidin-3-yl]-2-fluorophenyl}-1-cyclopropyl-6-fluoro-4-oxo-1,4-dihydro-3-quinolinecarboxylate, trifluoroacetic acid salt (94.5 mg, 0.158 moles) in 2.5 mL of pyridine at room 30 temperature was created via syringe with acetic anhydride (640 ΦL, 6.78 moles). The resulting amber solution was stirred under nitrogen at room temperature for 1 hour. TLC analysis indicated that the reaction was complete. The solution was cooled to

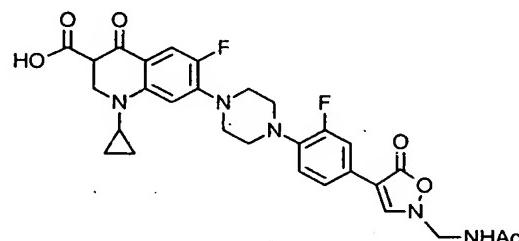
PATENT APPLICATION
28341/6304.NDV1

0EC and quenched with 0.6mL of methanol. After stirring under nitrogen at 0EC for 15 minutes and at room temperature for 20 minutes, the solvent was removed under reduced pressure. the crude product (112.2 mg) was chromatographed on a 14x80 mm Biotage column. The column was conditioned, loaded and eluted with 5% 4N NH₃- MeOH/CH₂Cl₂. Fractions 6-12 (5mL cuts) were combined, evaporated and dried under high vacuum at room temperature for 16 hours to yield Compound 10b (45.9 mg, 55%) as an off-white solid that melted at 186-187EC. MS(ESI+) for C₂₇H₂₅F₂N₃O₆: m/z 526.3 (M+H)⁺.

10

EXAMPLE 4

Synthesis of Quinolone-Isoxazolinones: 7-[2-Acetamidomethyl-2- hydroisoxazole-5-one-4-yl]-3-carboxy-1-cyclopropyl-1,4-dihydroquinoline-4-one (Compound 13).



15

The synthesis of Compound 13 includes several steps, as described below.

tert-Butyl 4-(4-formylphenyl)piperazinecarboxylate (Compound 28).

20

1-(t-Butoxycarbonyl)piperazine (1.86 g, 10 mmol), p-fluorobenzaldehyde 27 (1.24 g, 10 mmol), and K₂CO₃ (1.74 g, 11 mmol) in dry pyridine (10 mL) are stirred at reflux under a nitrogen atmosphere for 24 h. Most of the solvent is removed under vacuum and the residue is partitioned between EtOAc (150 mL) and water (100 mL). The aqueous layer is washed with EtOAc (2 x 50 mL) and the combined organic layers are sequentially washed with 3% aq. citric acid (2 x

PATENT APPLICATION

28341/6304.NDV1

100 mL), water (100 mL), brine (100 mL), and dried (MgSO_4). The solvent is removed under vacuum and the residue washed with hexanes and dried under vacuum. The product can be used as such, or further purified by silica gel column chromatography (dichloromethane -- MeOH gradient).

5

Ethyl (2E)-2-(4-{4-[(tert-butyl)oxycarbonyl]piperazinyl}phenyl)-3-hydroxyprop-2-enoate (Compound 29).

The synthesis is performed in an analogous manner to that described by Mahmood et al., *J. Org. Chem.*, 63, pgs. 3333-3336 (1998). Ethyl diazoacetate (6 mmol) in dichloromethane (40 mL) is added slowly (syringe pump, about 7 h) to a solution of aldehyde **27** (1.45 g, 5 mmol) and $[\text{Fe}(\text{Cp})(\text{CO})_2(\text{THF})]\text{BF}_4$ (0.5 mmol) in dichloromethane (60 mL) under a nitrogen atmosphere at 0 °C. The mixture is stirred at 0 °C for an additional 12 h, then ethyl ether (100 mL) is added. The resulting suspension is filtered through a plug of silica gel, the solvent is removed under vacuum, and the resulting Compound **27** is purified by column chromatography.

tert-Butyl 4-[4-(5-oxo-2-hydroisoxazol-4-yl)phenyl]piperazinecarboxylate (Compound 30).

Aqueous hydroxylamine (50%, 10 mmol) and Compound **29** (4 mmol) in MeOH (70 mL) and water (18 mL) are heated under reflux for 3 h. The MeOH is removed under vacuum, and the residue triturated with water. The precipitated Compound **30** is filtered, washed with cold water, and dried under vacuum.

tert-Butyl 4-(4-{2-[(acetylamino)methyl]-5-oxo-2-hydroisoxazol-4-yl}phenyl)piperazinecarboxylate (Compound 31).

N-(Hydroxymethyl)acetamide acetate (10 mmol) is added to Compound **30** (2 mmol) in dry dimethylformamide (18 mL), followed by K_2CO_3 (10 mmol). The mixture is stirred for 5 h, then poured into ice water. After about 18 h, the precipitated Compound **31** is filtered and dried under vacuum.

PATENT APPLICATION
28341/6304.NDV1

**N-{{[5-Oxo-4-(4-piperazinylphenyl)-2-hydroisoxazol-2-yl]methyl}acetamide
(Compound 12).**

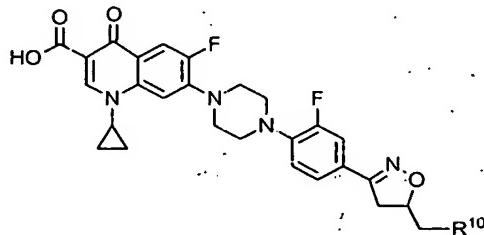
5 Trifluoroacetic acid (2 mL) is added to a solution of Compound 31 (1 mmol) in dichloromethane (5 mL). After 30 min, dichloromethane is removed under vacuum, and the product 12 is precipitated with ethyl ether, filtered, and dried under vacuum.

10 **7-[2-Acetamidomethyl-2- hydroisoxazole-5-one-4-yl]-3-carboxy-1-cyclopropyl--
1,4-dihydroquinoline-4-one(Compound 13).**

15 A mixture of Compound 12 (1.1 mmol) and 3-carboxy-1-cyclopropyl-7-chloro-6-fluoro-1,4-dihydroquinoline-4-one (1 mmol) in N-methylpyrrolidine-2-one (2.0 mL) and N-methylmorpholine (0.4 mL) is stirred at 110-130 °C for 48 h. The mixture is cooled to r.t., and a majority of the solvent is removed under vacuum. The residue is triturated with water, and the resulting precipitate is filtered, sequentially washed with excess water, THF, and ether, and dried under vacuum. Compound 13 is purified by preparative HPLC.

20

EXAMPLE 5



Synthesis of Quinolone-Isoxazolines:

7-(5-Acetamidomethyl-4,5-dihydroisoxazole-3-yl)-3-carboxy-1-cyclopropyl-1,4-dihydroquinoline-4-one (Compound 16, R¹⁰=NHAc).

5

The synthesis of the Compound 16 includes of several steps as described below.

tert-Butyl 4-[4-((hydroxyimino)methyl)phenyl]piperazinecarboxylate (Compound 32).

10

A mixture of aldehyde 28 (10 mmol) and hydroxylamine hydrochloride (15 mmol) in MeOH (20 mL) with pyridine (2 mL) is stirred at r.t. for 12-15 h. The solvent is removed under vacuum, and the residue is distributed between EtOAc (100 mL) and water (30 mL). The organic layer is washed with 2% aq. citric acid (2 x 30 mL), water (30 mL), brine (30 mL), and dried (MgSO₄). The solvent is evaporated and Compound 32 dried under vacuum.

15

tert-Butyl 4-[4-(chloro(hydroxyimino)methyl)phenyl]piperazinecarboxylate (Compound 33).

20

N-Chlorosuccinimide (12 mmol) is added portionwise with stirring to a solution of aldoxiome 32 (10 mmol) in dichloromethane (50 mL) with pyridine (10 mL) at 0 °C. The mixture is stirred at this temperature for 3 h, and then 1-2 h at r.t. The solvent is removed under vacuum, the residue is triturated with ice water, and the precipitated Compound 33 is sequentially washed with ice water, cold 3% aq. citric acid, water, and dried under vacuum.

tert-Butyl 4-(4-{5-[(acetylamino)methyl]-4,5-dihydroisoxazol-3-yl}phenyl)piperazinecarboxylate (Compound 34).

5 Triethylamine (7.5 mmol) is added dropwise with stirring to the mixture of N-acetylallylamine (7.5 mmol) and Compound 33 (5 mmol) in dichloromethane (20 mL). The mixture is stirred at r.t. overnight. The solvent is removed under vacuum, and the residue is distributed between EtOAc (100 mL) and water (30 mL). The organic layer is sequentially washed with 2% aq. citric acid (2 x 10 30 mL), water (30 mL), brine (30 mL), and dried (MgSO_4). The solvent is evaporated and Compound 34 is purified by silica gel column chromatography (eluent: dichloromethane – MeOH gradient).

15 N-{[3-(4-Piperazinylphenyl)-4,5-dihydroisoxazol-5-yl]methyl}acetamide (Compound 14).

20 Trifluoroacetic acid (2 mL) is added to a solution of Compound 34 (1 mmol) in dichloromethane (5 mL). After 30 min, dichloromethane is removed under vacuum, and Compound 14 is precipitated with ethyl ether, filtered, and dried under vacuum.

7-(5-Acetamidomethyl-4,5-dihydroisoxazole-3-yl)-3-carboxy-1-cyclopropyl-1,4-dihydroquinoline-4-one (Compound 16).

25 A mixture of Compound 14 (1.1 mmol) and 3-carboxy-1-cyclopropyl-7-chloro-6-fluoro-1,4-dihydroquinoline-4-one (1 mmol) in NMP (2.0 mL) and NMM (0.4 mL) is stirred at 110-130 °C for 48 h. The mixture is cooled to r.t., and a majority of the solvent is removed under vacuum. The residue is triturated with water, the resulting precipitate is filtered, washed with excess water, THF, ether, and dried under vacuum. Compound 16 is purified by preparative HPLC. Optionally, individual 5-(R)- and 5-(S)-enantiomers (resulting from the racemic center in the

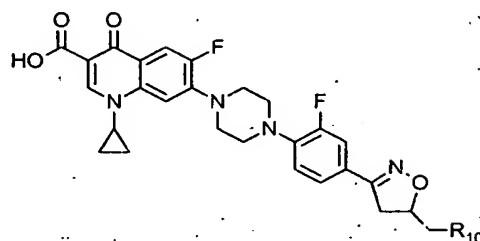
PATENT APPLICATION

28341/6304.NDV1

isoxazoline group) are separated by HPLC using chiral phase (such as Chiralpack AD column).

EXAMPLE 6

5



Synthesis of Quinolone-Isoxazolines: 7-(5-Hydroxymethyl-4,5-dihydroisoxazol-3-yl)-3-carboxy-1-cyclopropyl-1,4-dihydroquinoline-4-one (Compound 17, R¹⁰=OH).

10

The synthesis of Compound 17 includes several steps as described below.

15

tert-Butyl 4-(4-{5-hydroxymethyl-4,5-dihydroisoxazol-3-yl}phenyl)piperazinecarboxylate (Compound 35).

20

Triethylamine (7.5 mmol) is added dropwise with stirring to a mixture of allyl alcohol (7.5 mmol) and Compound 32 (5 mmol) in dichloromethane (20 mL). The resulting mixture is stirred at r.t. overnight. The solvent is removed under vacuum and the residue is partitioned between EtOAc (100 mL) and water (30 mL). The organic layer is washed with 2% aq. citric acid (2 x 30 mL), water (30 mL), brine (30 mL), and dried (MgSO₄). The solvent is evaporated, and Compound 35 is purified by silica gel column chromatography (eluent: dichloromethane – MeOH gradients).

PATENT APPLICATION
28341/6304.NDV1

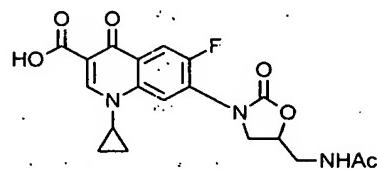
3-(4-Piperazinylphenyl)-4,5-dihydroisoxazol-5-ylmethanol (Compound 15).

5 Trifluoroacetic acid (2 mL) is added to a solution of Compound 35 (1 mmol) in dichloromethane (5 mL). After 30 min, dichloromethane is removed under vacuum, and Compound 15 is precipitated with ethyl ether, filtered, and dried under vacuum.

10 **7-(5-Hydroxymethyl-4,5-dihydroisoxazole-3-yl)-3-carboxy-1-cyclopropyl-1,4-dihydroquinoline-4-one (Compound 17).**

15 A mixture of Compound 15 (1.1 mmol) and 3-carboxy-1-cyclopropyl-7-chloro-6-fluoro-1,4-dihydroquinoline-4-one (1 mmol) in NMP (2.0 mL) and NMM (0.4 mL) is stirred at 110-130 °C for 48 h. The mixture is cooled to r.t., and a majority of the solvent is removed under vacuum. The residue is triturated with water, and the resulting precipitate filtered, sequentially washed with excess water, THF, ether, and dried under vacuum. Compound 17 is purified by preparative HPLC. Optionally, individual 5-(R)- and 5-(S)-enantiomers (resulting from the racemic center in isoxazoline group) are separated by HPLC using chiral phase (such as Chiralpack AD column).

EXAMPLE 7



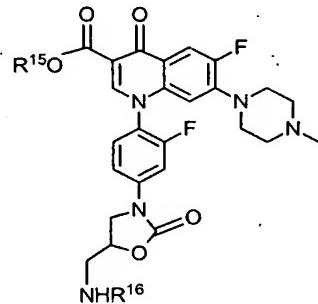
PATENT APPLICATION

28341/6304.NDV1

7-[5-(S)-Acetamidomethyloxazolidine-2-one-3-yl]-3-carboxy-1-cyclopropyl-6-fluoro-1,4-dihydroquinoline-4-one (Compound 19).

Compound 19 is prepared according to the General Procedure
5 Synthesis of Quinolone-7-yl Linked Oxazolidinones from 1-acetyl-3-amino-2-(S)-oxypropylamine (2 mmol) using 3-carboxy-1-cyclopropyl-7-chloro-6-fluoro-1,4-dihydroquinoline-4-one (0.282 g, 1 mmol). The first reaction step is performed at 120 – 130 °C for 48 h. The mixture is cooled to room temperature (r.t.), and a majority of the solvent removed under vacuum. The resulting amino alcohol intermediate is
10 purified by silica gel column chromatography (eluent: dichloromethane -- MeOH). Cyclization is performed by dissolving the amino alcohol (1 mmol) in an aprotic organic solvent (e.g., THF or NMP) (2-5 mL), in the presence of carbonyldiimidazole (1.1 mmol) and adding imidazole (1.0 mmol). The mixture is stirred at 20 - 40 °C for
15 1 - 3 h. The solvent is removed under vacuum, and the product is purified by silica gel column chromatography (eluent: dichloromethane -- MeOH).

EXAMPLES 8-10



N¹-Linked Quinolone-Oxazolidinones: 3-Carboethoxy-1-[4-(5-(S)-acetamidomethyloxazolidine-2-one-3-yl)-2-fluorophenyl]-7-(4-methylpiperazine-1-yl)-6-fluoro-1,4-dihydroquinoline-4-one (Compound 24, R¹⁵=Et, R¹⁶=Ac), 3-Carboxy-1-[4-(5-(S)- aminomethyloxazolidine-2-one-3-yl)-2-fluorophenyl]-7-(4-methylpiperazine-1-yl)-6-fluoro-1,4-dihydroquinoline-4-one (Compounds 25, R¹⁵=H, R¹⁶=H), and 3-Carboxy-1-[4-(5-(S)- acetamidomethyloxazolidine-2-one-3-

PATENT APPLICATION

28341/6304.NDV1.

yl)-2-fluorophenyl]-7-(4-methylpiperazine-1-yl)-6-fluoro-1,4-dihydroquinoline-4-one (**Compound 26**, R¹⁵=H, R¹⁶=Ac).

The synthesis of the above compounds was performed in several steps
5 by the procedure described below.

Ethyl (2Z)-3-[{4-[(acetylamino)methyl]-2-oxo(1,3-oxazolidin-3-yl)}-2-fluorophenyl]amino]-2-[(2,4,5-trifluorophenyl)carbonyl]prop-2-enoate (Compound 22).

10

15

20

25

Triethylamine (2.09 mL, 15 mmol) was added with stirring to a solution of 2,4,5-trifluorobenzoyl chloride (1.95 g, 10 mmol) and ethyl 3-dimethylaminoacrylate **20** (1.43 g, 10 mmol) in 1,4-dioxane (20 mL). The mixture was stirred at r.t. overnight, and solvent was removed under vacuum. The residue was dissolved in ethyl ether (100 mL) and hexanes (20 mL), and the resulting mixture was washed with water (2 x 100 mL), 2% aq. citric acid (2 x 100 mL), brine (100 mL), 2.5% aq. NaHCO₃ (2 x 100 mL), and brine (100 mL). The organic layer was dried (MgSO₄), and the solvents removed under vacuum to provide ethyl-2-(dimethylamino)methylene-2-(2,4,5-trifluorobenzoyl)acetate **21** as a light orange oil (Yield 2.55 g (85%). MS (m/z): 302 [M+H]⁺. R_t 4.9 min.). A solution of Compound **21** (0.301 g, 1.0 mmol) and 5-(S)-acetamidomethyl-3-(4-amino-3-fluorophenyl)-oxazolidine-2-one (0.267 g, 1.0 mmol) in EtOH (9 mL) was agitated at 40 °C for 16 h. The solvent was removed under vacuum, and the resulting residue dissolved in ethyl ether (75 mL). The ether solution was washed with 3% aq. citric acid (2 x 40 mL), brine (40 mL), and dried (MgSO₄). The solvent was removed under vacuum to afford Compound **22** as a thick light-orange oil. Yield 0.50 g (90%). MS (m/z): 524 [M+H]⁺, R_t 5.5 min.

PATENT APPLICATION
28341/6304.NDV1

1-(4-{5-[(acetylamino)methyl]-2-oxo(1,3-oxazolidin-3-yl)}- 2-fluorophenyl)-6,7-difluoro-4-oxohydroquinoline-3-carboxylate (Compound 23).

5 Compound 22 (0.40 g, 0.76 mmol) was heated under a nitrogen atmosphere with 3% diazabicyclo[4.4.0]undec-2-ene in N-methylpyrrolidine-2-one (4 mL) for 30 min. at 80 °C. Most of the solvent was removed under a high vacuum, and the residue partitioned between ethyl acetate (EtOAc) (50 mL) and water (30 mL). The organic layer was sequentially washed with water (30 mL), 3% aq. citric acid (2 x 30 mL), water (30 mL), brine (30 mL), and dried (MgSO_4). The solvent was removed under vacuum and the residue washed with an excess of ethyl ether to afford Compound 23 as orange crystals. Yield 0.275 g (72%). MS (m/z): 504 $[\text{M}+\text{H}]^+$. R_t 4.5 min.

15 **3-Carboethoxy-1-[4-(5-(S)- acetamidomethyloxazolidine-2-one-3-yl)-2-fluorophenyl]-7-(4-methylpiperazine- 1-yl)-6-fluoro-1,4-dihydroquinoline-4-one (Compound 24) and 3-Carboxy-1-[4-(5-(S)- aminomethyloxazolidine-2-one-3-yl)-2-fluorophenyl]- 7-(4-methylpiperazine- 1-yl)-6-fluoro-1,4-dihydroquinoline-4-one (Compound 25).**

20 A solution of Compound 23 (0.05 g, ca. 0.1 mmol) and 1-methylpiperazine (0.04 mL, 0.34 mmol) in N-methylpyrrolidine-2-one was heated at 80 °C for 18 h. A majority of the solvent was removed under a high vacuum, and the resulting crude Compound 24 heated with 6N aq. HCl (3 mL) in a sealed vial for 4h at 25 80 °C. The solvent was evaporated under vacuum to about 2 mL, and amine 25 purified by preparative RP HPLC. MS (m/z): 514 $[\text{M}+\text{H}]^+$. R_t 3.5 min.

30 **3-Carboxy-1-[4-(5-(S)- acetamidomethyloxazolidine-2-one-3-yl)-2-fluorophenyl]-7-(4-methylpiperazine- 1-yl)-6-fluoro-1,4-dihydroquinoline-4-one (Compound 26).**

PATENT APPLICATION
28341/6304.NDV1

Acetic anhydride (0.05 mL) was added to a suspension of amine 25 (0.01 g, 0.016 mmol) with poly(vinylpyridine) (0.1 g) in MeCN (3 mL). The mixture was agitated for 30 min at r.t. The supernatant was filtered, the separated resin washed with MeOH (2 x 3 mL), and the combined filtrates evaporated under vacuum. 5 The resulting solid was recrystallized from a minimum of MeOH to afford Compound 26 as white crystals. Yield 6.9 mg (64%). MS (m/z): 556 [M+H]⁺. R_t 3.7 min.

As shown in Table 1, the compounds of Example 1, 2 and 3 demonstrated a potent antibacterial activity.

10

Table 1— Antibacterial Activity of Selected Examples

Example No.	MIC(:g/mL)					
	<i>E.faecalis</i> UC9217	<i>S. aureus</i> UC9218	<i>S. pneumoniae</i> UC9912	<i>H. influenzae</i> UC30063	<i>M. catarrhalis</i> UC30607	<i>E. coli</i> UC6674
1	0.25	0.5	0.125	8	1	16
2	0.5	1	0.25	4	2	16*
3	8	2	4	0.5	4	4

The compounds of the invention can be used for the treatment or prevention of infectious disorders caused by variety of bacterial organisms. Examples include Gram positive and Gram negative aerobic and anaerobic bacteria, including 15 *Staphylococci*, for example *S. aureus*; *Enterococci*, for example *E. faecalis*; *Streptococci*, for example *S. pneumoniae*; *Haemophilis*, for example *H. influenza*; *Moraxella*, for example *M. catarrhalis*; and *Escherichia* for example *E. coli*. Other examples include *Mycobacteria*, for example *M. tuberculosis*; intercellular microbes, 20 for example *Chlamydia* and *Rickettsiae*; and *Mycoplasma*, for example *M. pneumoniae*.

PATENT APPLICATION

28341/6304.NDV1

It will be apparent to those skilled in the art that various modifications and variations can be made in the present invention without departing from the scope or spirit of the invention. Other embodiments of the invention will be apparent to those skilled in the art from consideration of the specification and practice of the invention disclosed herein. It is intended that the specification and examples be considered as exemplary only, with a true scope and spirit of the invention being indicated by the following claims.